

1 agggagagggc agtgaccatg aaggctgtgc tgcctgccc tttgatggca
 51 ggcttggccc tgcagccagg cactgcccgt ctgtgctact cctgcaaagc
 101 ccaggtgagc aacgaggact gccctgcaggt ggagaactgc acccagctgg
 151 gggagcagtg ctggaccgcg cgcaaccgcg cagtggcct cctgaccgtc
 201 atcagcaaag gctgcagctt gaactgcgtg gatgactcac aggactacta
 251 cgtgggcaag aagaacatca cgtgctgtga caccgacttg tgcaacgcca
 301 gcgggggccc tgcctgcag ccggctgccg ccatcctgc gctgctccct
 351 gcactcggcc tgcctgctctg gggaccggc cagctatagg ctctgggggg
 401 ccccgctgca gccacactg ggtgtgtgc cccaggcctt tgtgccactc
 451 ctacagaac ctggcccagt gggagcctgt cctggctcct gaggcacatc
 501 ctaacgcaag ttgaccatg tatgttgca cccctttcc cnaaccctg
 551 acctcccat gggcctttc caggatccn accnggcaga tcagtittag
 601 tganacanat ccgctgcag atggccctc caacnnttn tgttgnntn
 651 tccatggccc agcatttcc accttaacc ctgtgtcag gcacttntc
 701 cccaggaag cctccctgc ccacccan tatgaattga gccaggttg
 751 gtccgtgtg tccccgcac ccagcagggg acaggcaatc aggagggccc
 801 agtaagggc gagatgaagt ggactgagta gaactggagg acagagtg
 851 acgtgagtc ctgggagtc ccagagatgg ggcctggagg cctggaggaa
 901 gggggccaggc ctacattg tggggtccc gaatggcagc ctgagcaccg
 951 cgtaggccct taataaacac ctgtggata agccaaaaa aaaaaaaa

FIGURE 1A

[illegible]

11

1 ATGAAGACAGT TTTT TTTATCCTGCTGGCCACCTACTTAGCCCTGCATCCAGGTGCTGCT 60
 TACTTCTGTCAAAAAAATAGGACGACCGGTGGATGAATCGGGACGTAGGTCCACGACGA
 M K T V F F I L L A T Y L A L H P G A A -
 CTGCAGTGCTATTTCATGCACAGCACAGATGAACAACAGAGACTGTCTGAATGTACAGAAC
 61 120
 GACGTCACGATAAGTACGTGTCGTGCTACTTGTGTCTCTGACAGACTTACATGTCTTG
 L Q C Y S C T A Q M N N R D C L N V Q N -
 TGCAGCCTGGACCAGCACAGTTGCTTTACATCGCGCATCCGGGCCATTGGACTCGTGACA
 121 180
 ACGTCGGACCTGGTGTGTCACGAAATGTAGCGCGTAGGCCCGGTAACCTGAGCACTGT
 C S L D Q H S C F T S R I R A I G L V T -
 GTTATCAGTAAGGGCTGCAGCTCACAGTGTGAGGATGACTCGGAGAACTACTATTTGGGC
 181 240
 CAATAGTCATTCCCGACGTCGAGTGTCACTCTACTGAGCCTCTTGATGATAAACCCG
 V I S K G C S S Q C E D D S E N Y Y L G -
 AAGAAGAACATCACGTGCTGCTACTCTGACCTGTGCAATGTCAACGGGGCCACACCTG
 241 300
 TTCTTCTTGTAGTGCACGACGATGAGACTGGACACGTTACAGTTGCCCCGGGTGTGGGAC
 K K N I T C C Y S D L C N V N G A H T L -
 AAGCCACCCACCACTGGGGCTGCTGACCGTGCTCTGCAGCCTGTTGCTGTGGGGCTCC
 301 360
 TTCGGTGGGTGGTGGGACCCGACGACTGGCACGAGACGTCCGACAACGACACCCCGAGG
 K P P T T L G L L T V L C S L L L W G S -
 AGCCGTCGTAGGCTCTGGGAGAGCCTACCATAGCCCGATTGTGAAGGGATGAGCTGCAC
 361 420
 TCGGCAGACATCCGAGACCCTCTCGGATGGTATCGGGCTAACACTTCCCTACTCGACGTG
 S R L *
 TCCACCCCAACCCACACAGG
 421 441
 AGGTGGGGTGGGGTGTGTCC

FIGURE 2

[Faint, illegible handwritten notes or bleed-through from the reverse side of the page.]

FIGURE 3

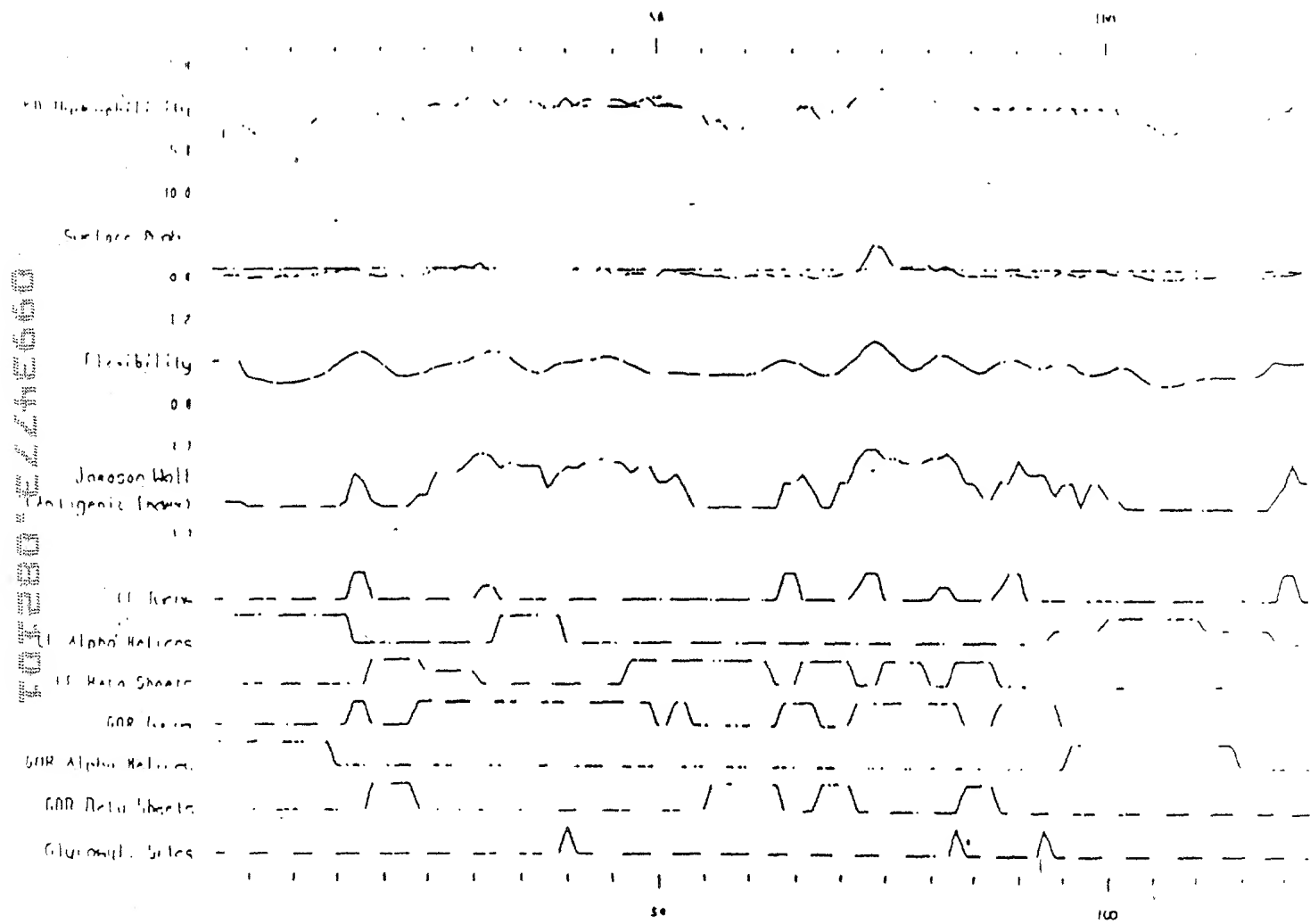


FIGURE 4

099473 0824
000000 000000

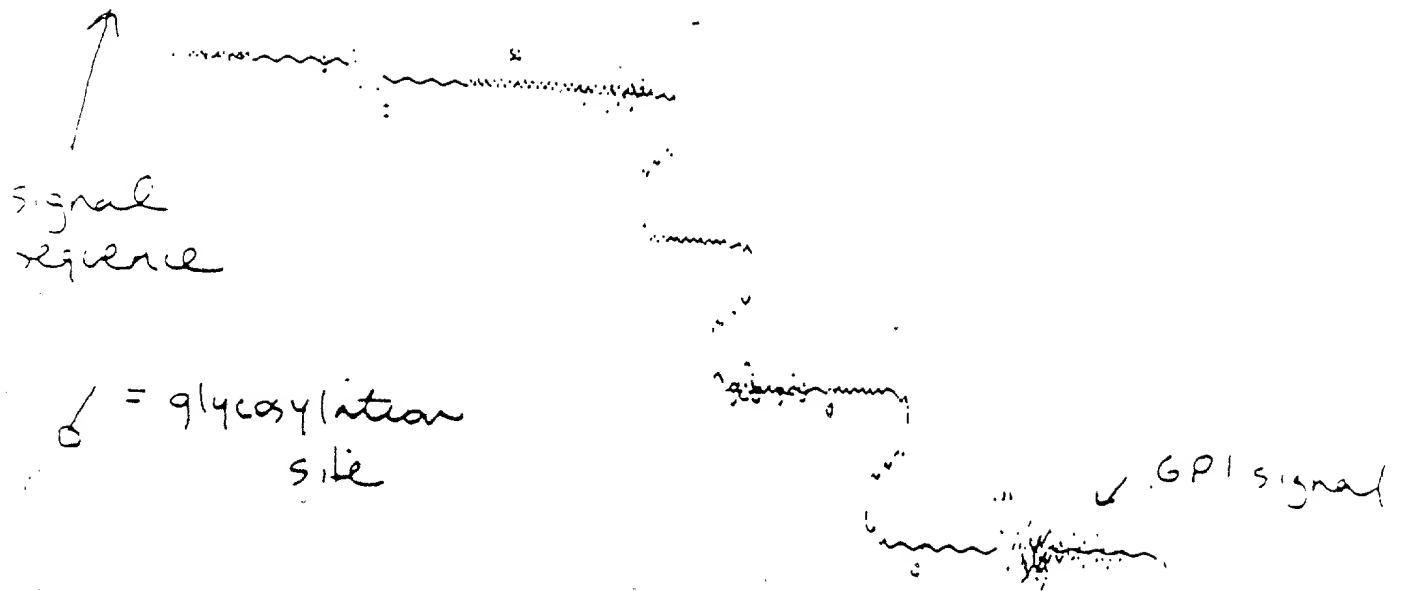


FIGURE 5

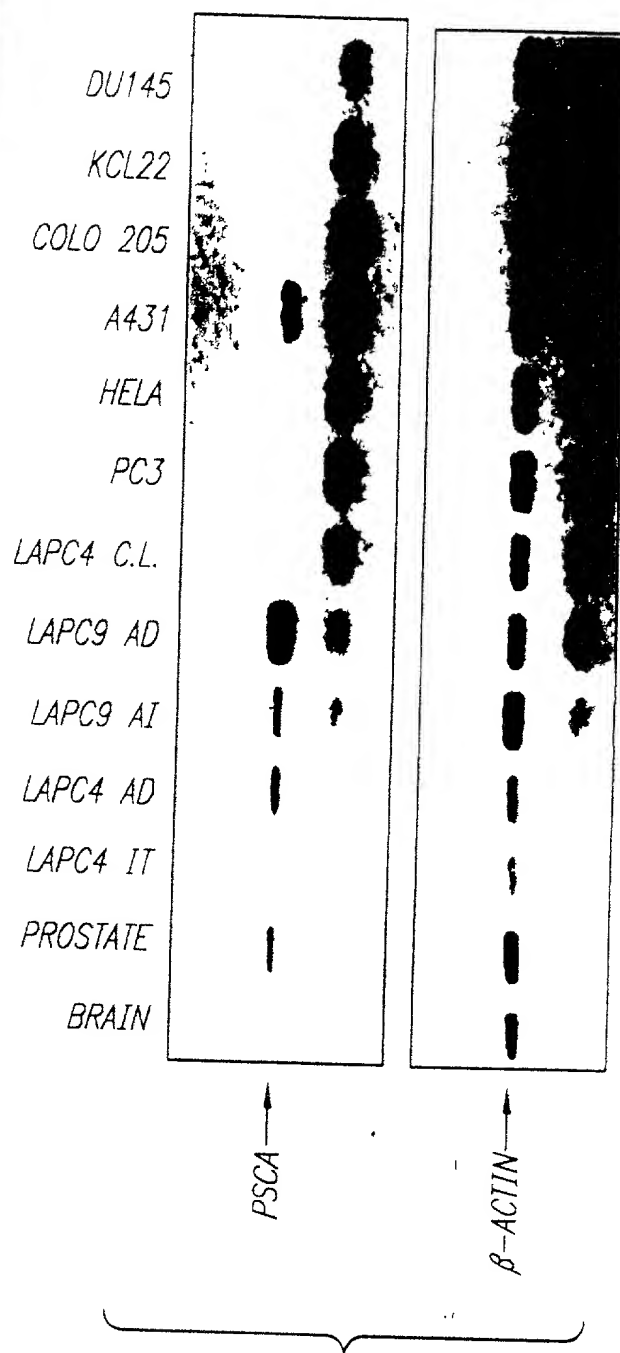
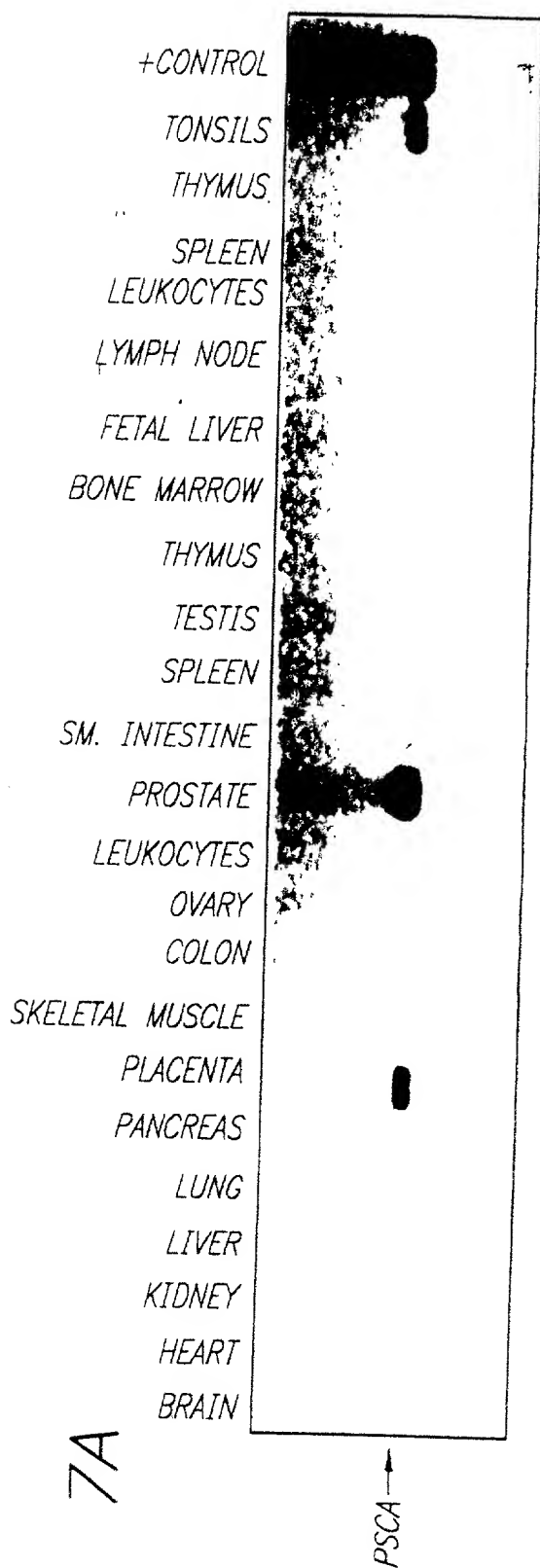
Western ASCA
 Superoxide 80mg/ml
 Normal tissue
 1hr exp



1G8
 1:100

prostate (Kumar)
 prostate (Baker)
 prostate (Glick)
 Bladder (Kumar)
 Bladder (Glick)
 Bladder (Rob)
 Kidney (Nelson)
 Kidney (Waller)
 Testis
 Sm. Intest.

LA PCR

FIGURE 6



Legend:  untranslated region of PSCA
 translated region of PSCA

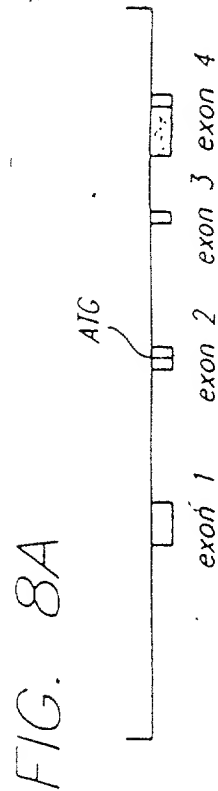


FIG. 8B

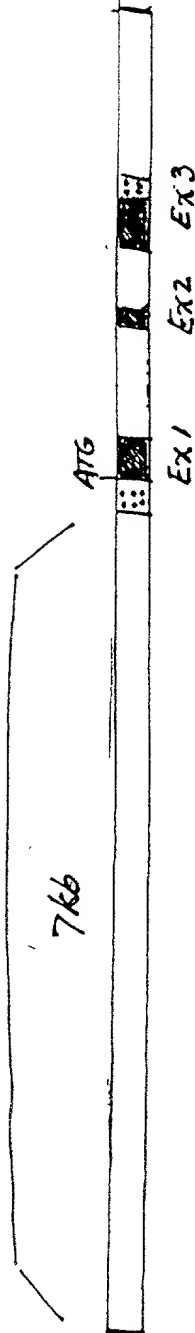
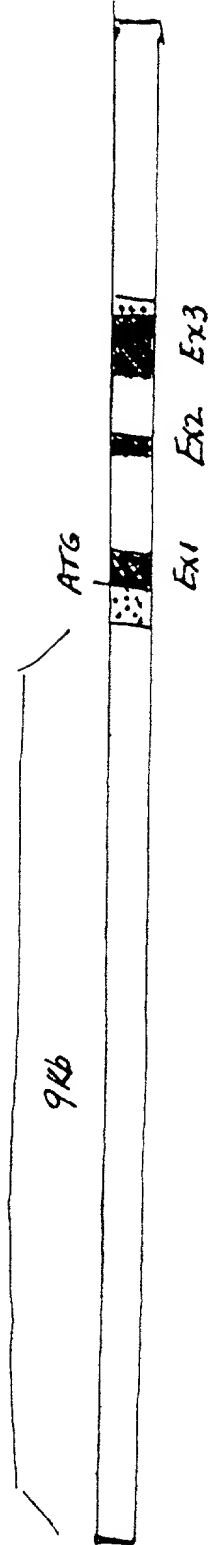


FIG. 8C



human PSCA

FIGURE 8

human PSCA

PSCA / PSA Expression in Benign
Prostate vs. Prostate Cancer Xenograft

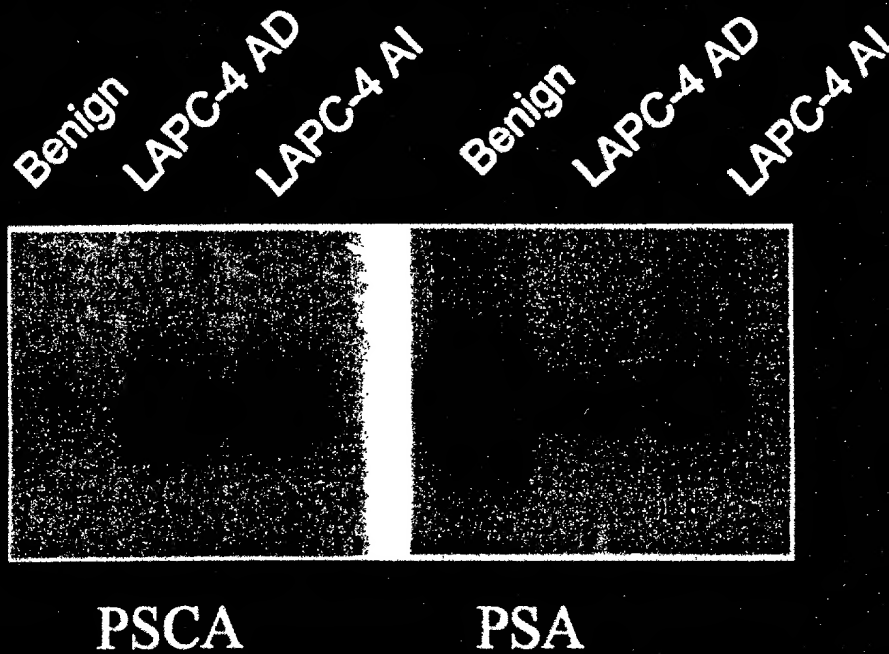


FIGURE 9A

~1kb

PANCREAS
KIDNEY
SKELETAL MUSCLE
LIVER
LUNG
PLACENTA
BRAIN
HEART

HERAL LEUKOCYTES
COLON
SMALL INTESTINE
OVARY
TESTIS
PROSTATE
THYMUS
SPLEEN

PSCA

FIG. 9B

404280 224650

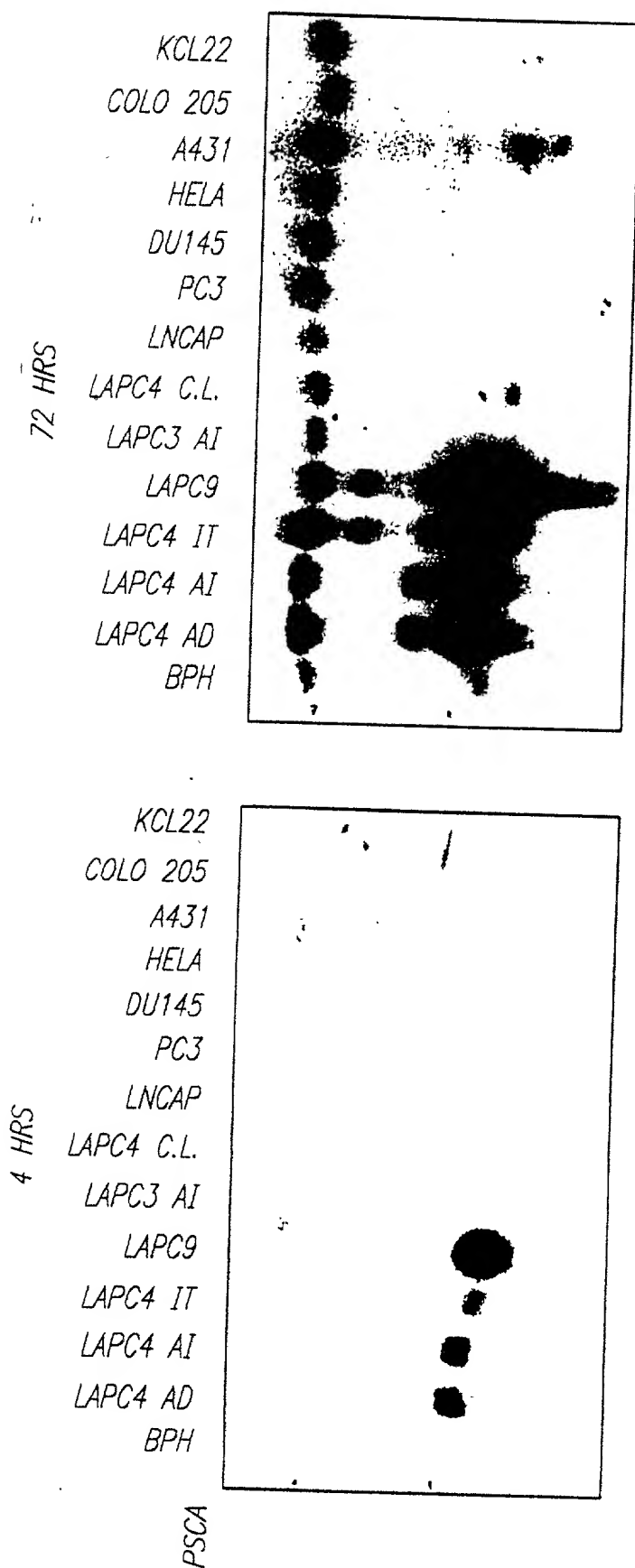
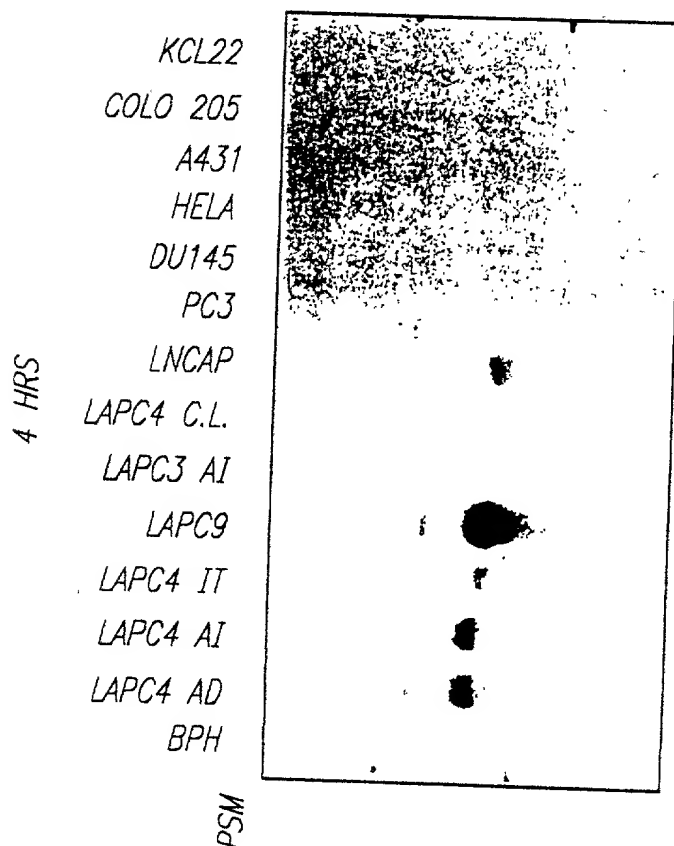
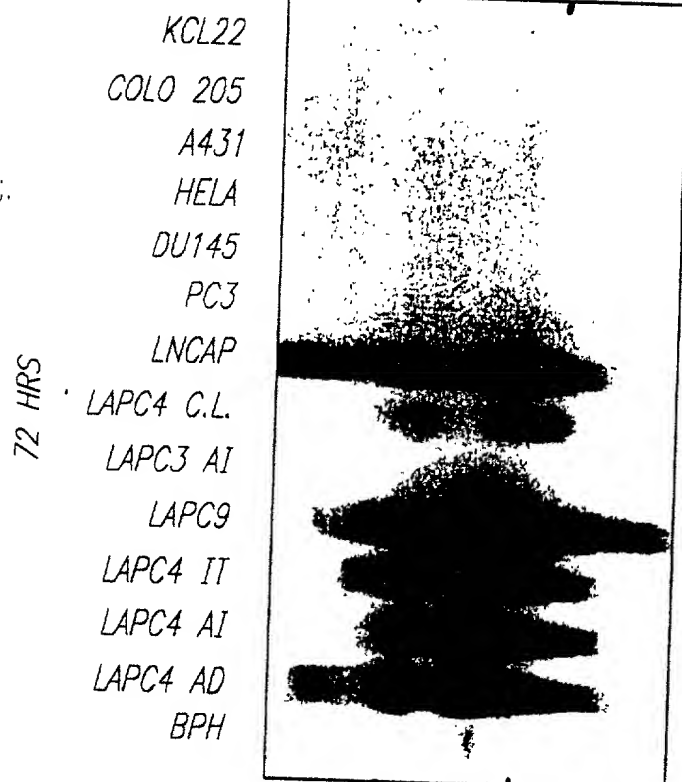


FIG. 10-1



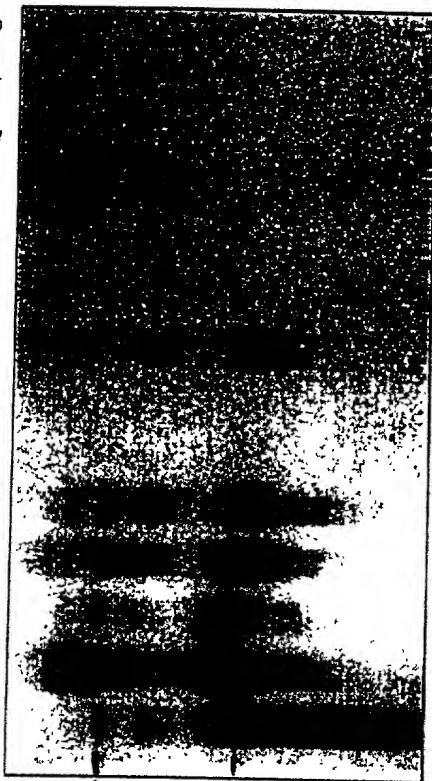
PSM

FIG. 10-2

707280 6/24/66

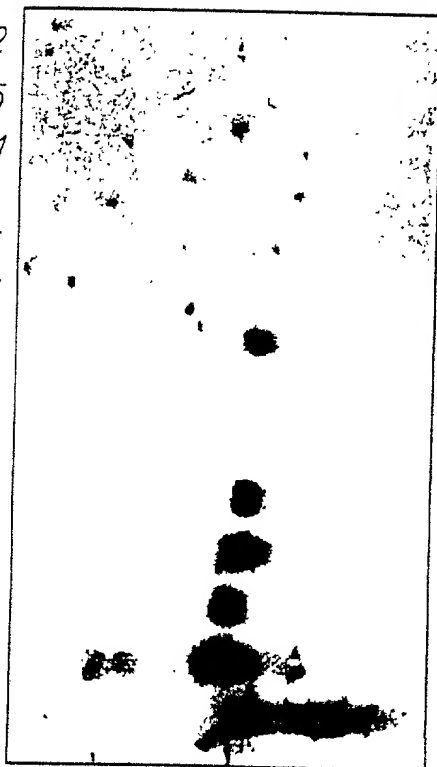
72 HRS

KCL22
COLO 205
A431
HELA
DU145
PC3
LNCAP
LAPC4 C.L.
LAPC3 AI
LAPC9
LAPC4 IT
LAPC4 AI
LAPC4 AD
BPH

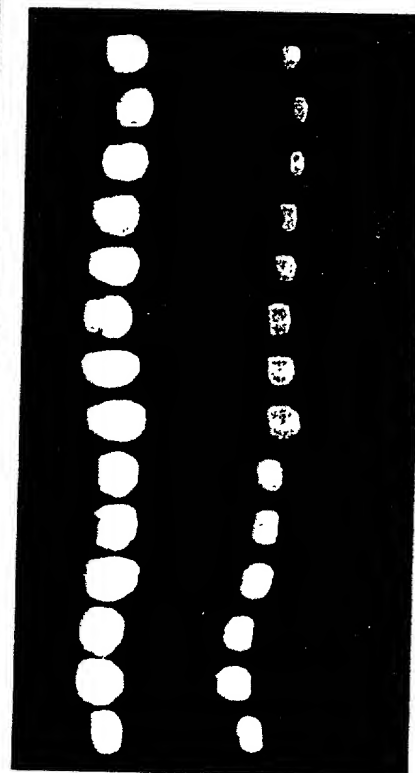


4 HRS

KCL22
COLO 205
A431
HELA
DU145
PC3
LNCAP
LAPC4 C.L.
LAPC3 AI
LAPC9
LAPC4 IT
LAPC4 AI
LAPC4 AD
BPH



PSA



ETBR

FIG. 10-3

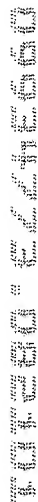
[illegible]

FIG. 11B

707280" E/24E660

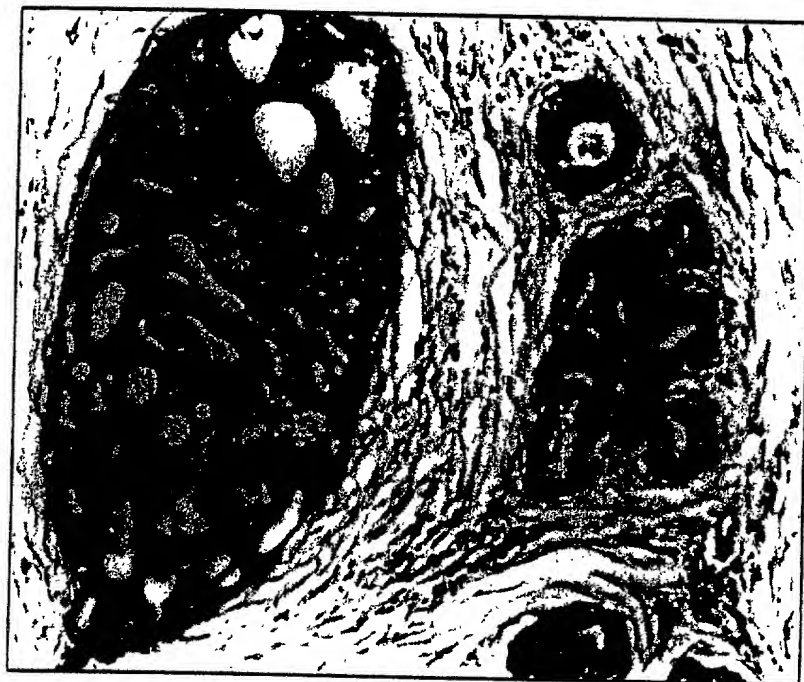


FIG. 11C

101.233 2248660

FIG. 12A

O GLYCOSIDASE
N GLYCOSIDASE F
CONTROL

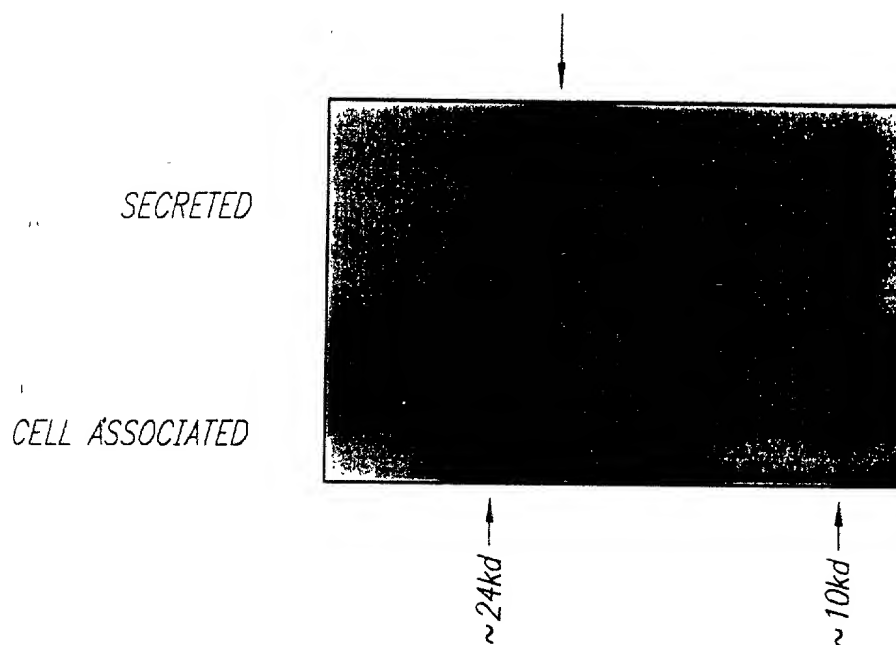
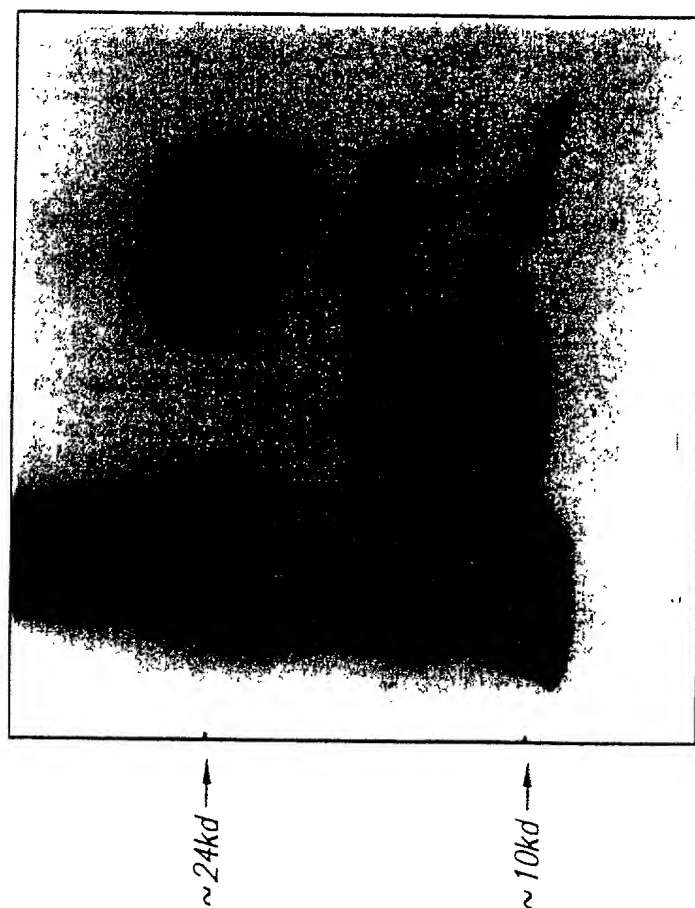


FIG. 12B

407230" E 242.660

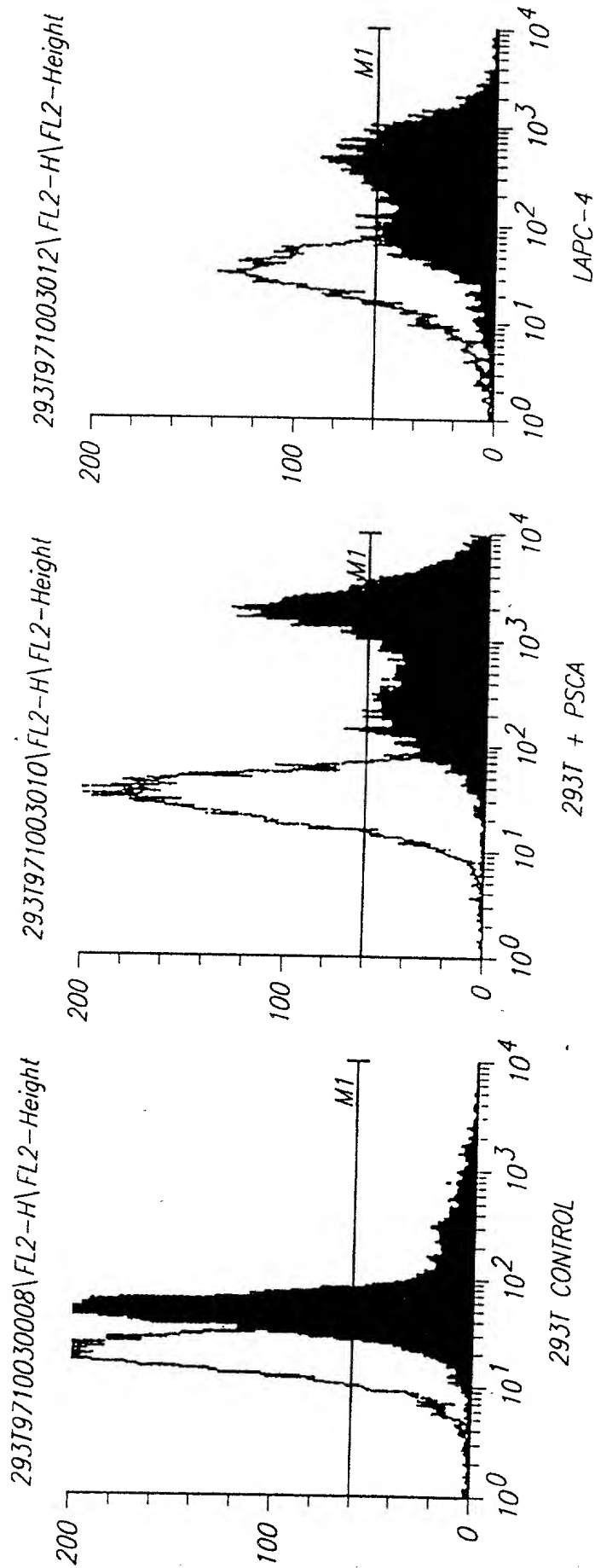


FIGURE 12C

PSCA Maps to Chromosome 8q24.2



Fluorescent
in Situ Hybridization
Analysis of PSCA

FIGURE 13

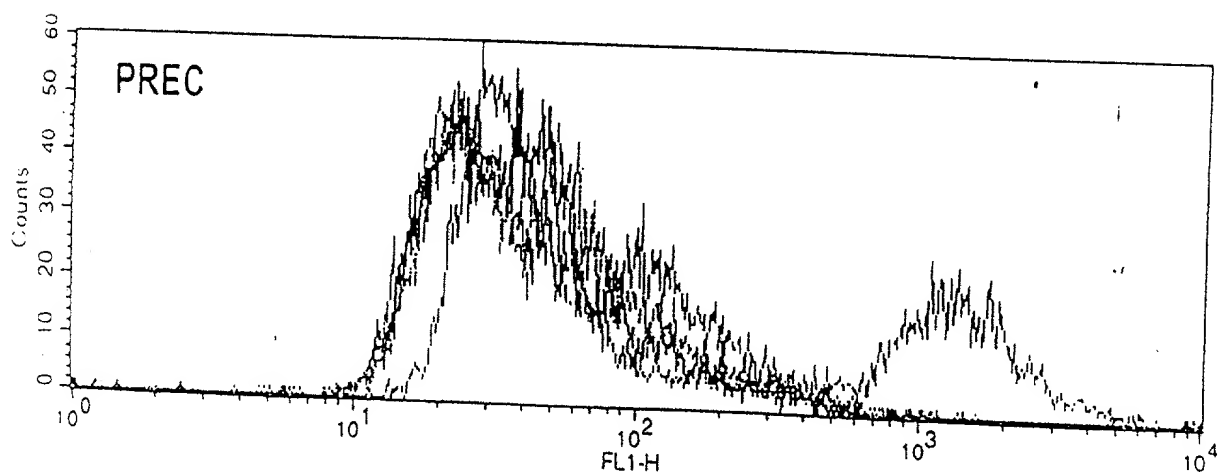
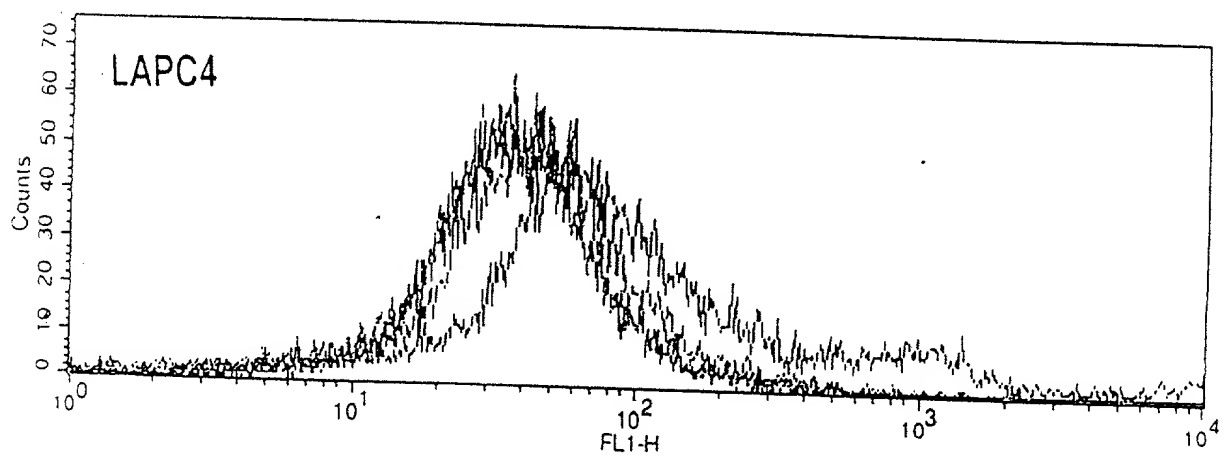
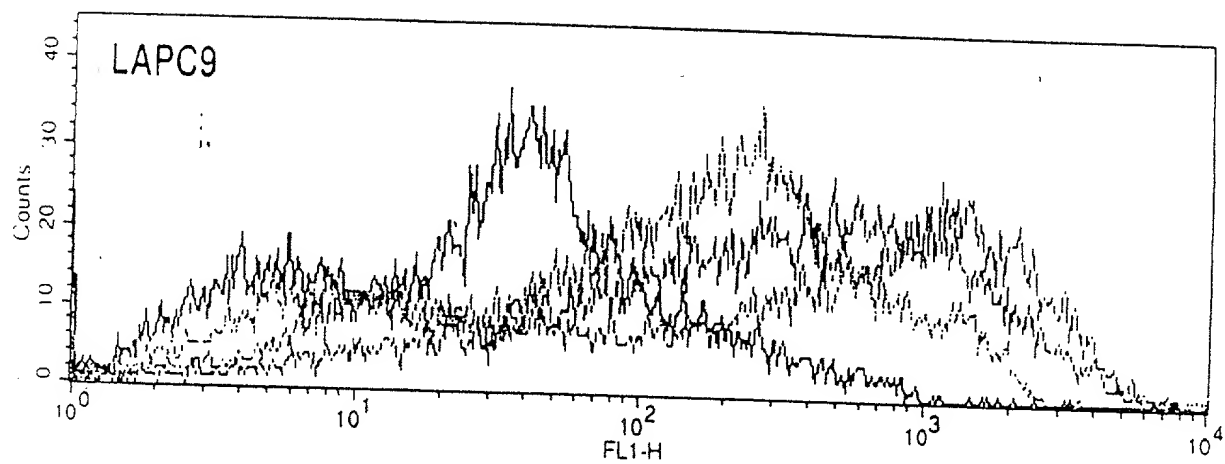


FIGURE 14

A

Epitope map

mAb	Isotype	FL (18-98)	N (2-50)	M (46-109)	C (85-123)
1G8	IgG1 k	2.039	0.007	0.628	0.000
2H9	IgG1 k	1.318	0.863	0.032	0.021
3C5	IgG2a k	2.893	1.965	0.016	0.005
3E6	IgG3 k	0.328	0.024	0.069	0.370
4A10	IgG2a k	2.039	1.315	0.000	0.014
2A2	IgG2a k	1.366	0.733	0.010	0.003
3G3	IgG2a k	2.805	1.731	0.004	0.000

B

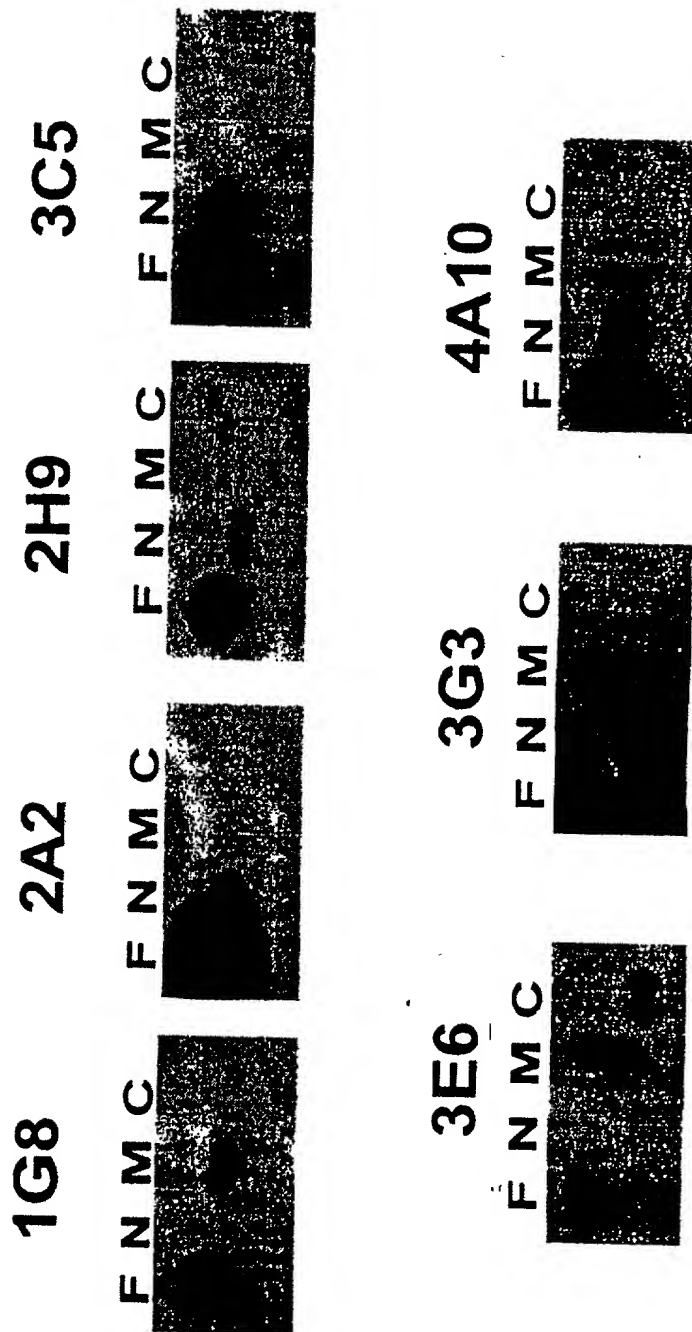


FIGURE 15

[Faint, illegible handwritten notes or bleed-through from the reverse side of the page.]

hSCA-2
hPSCA
mPSCA

21 M C F S C C Q A S N * C L V N *
21 Q C G S C T K O M N P D C L N V N *

41 C S C T R I R A A G L T
41 C S C T R I R A A G L T

61 V - - - - - L S K I C S C
61 V - - - - - L S K I G C S C C W D D S
61 V - - - - - I S K I G C S C C W D D S

81 V N T D V A S M S I C C C 2 S F L C N *
76 D Y G K K - N E I T C C T D L C N *
76 N Y L G K K - N E I T C C V S D L C N *

101 S A D G C R A L L L L L
95 S A H A U C P A L L L L
95 N G H T E F P P L L L

121 S V P A C C T E
115 S P L L
115 S P L L

(Reiter, R.E., et al., 1997. *PNAS*)

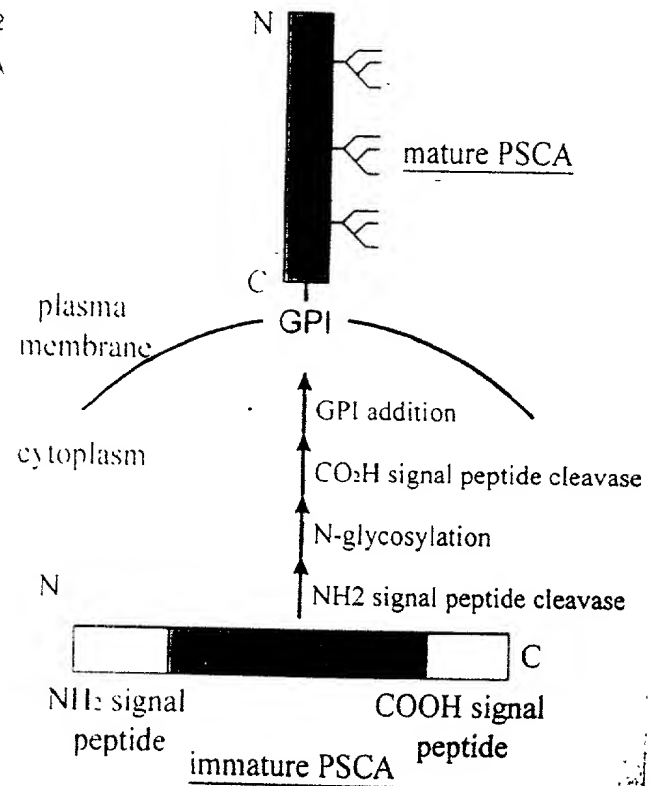
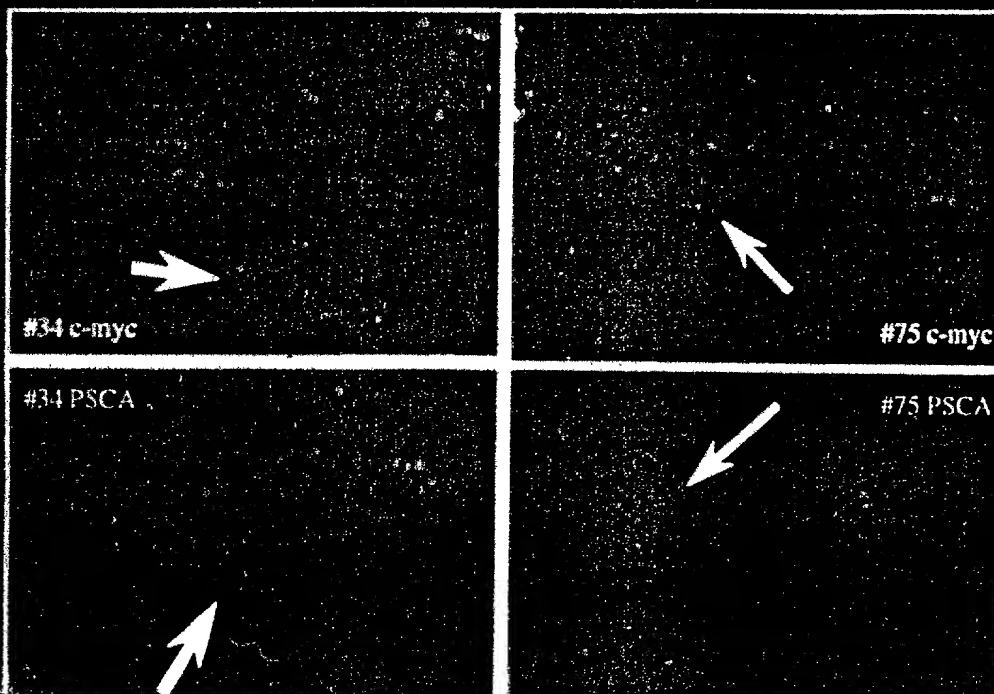


FIGURE 16

FISH Analysis of PSCA and c-myc in Prostate Cancer

Gain Chromosome 8

Amplification



R. Jenkins

FIGURE 17

10/20/2000 10:44:00

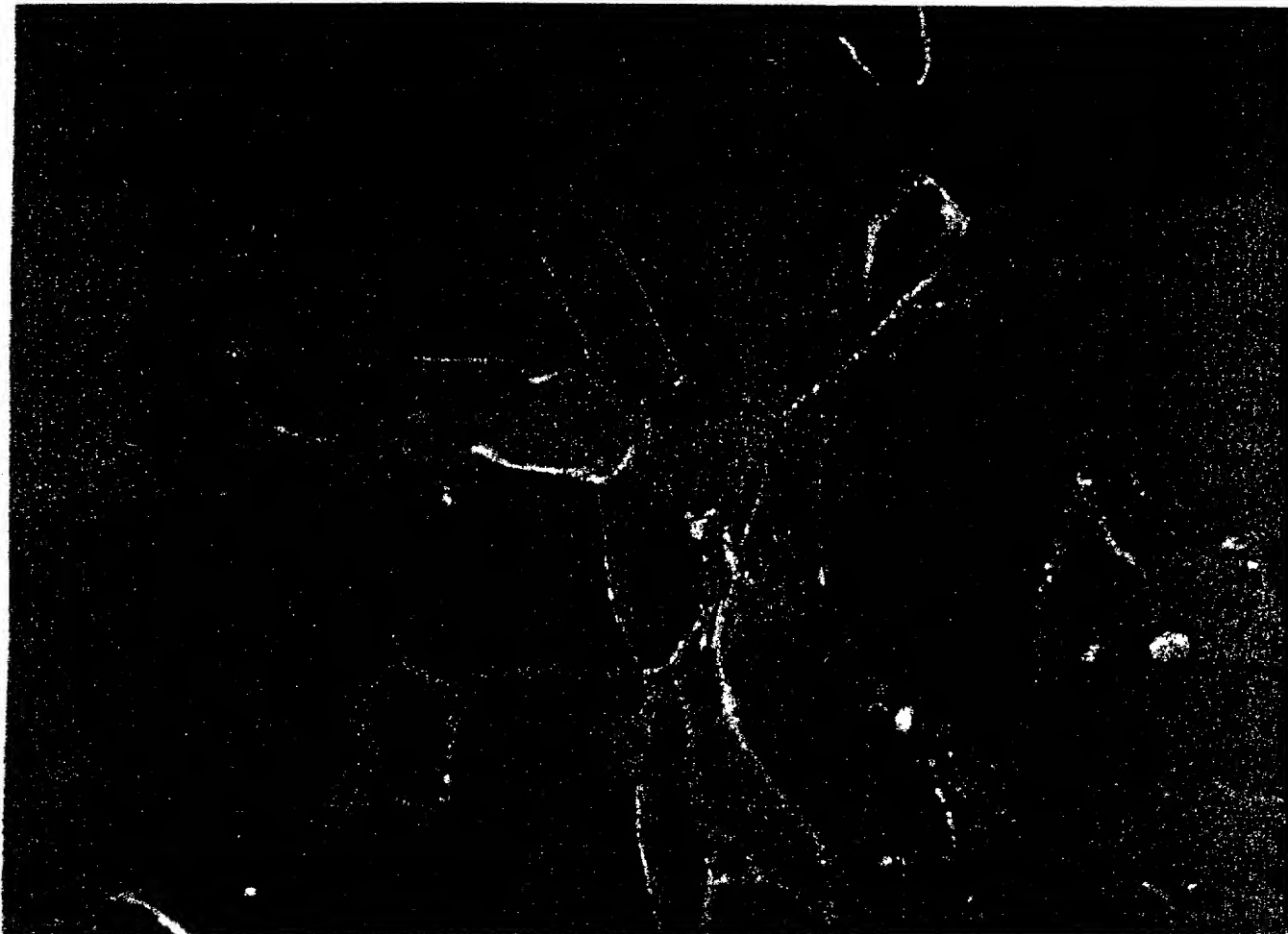


FIGURE 18

1. 2. 3. 4. 5. 6. 7. 8. 9. 10. 11. 12. 13. 14. 15. 16. 17. 18. 19. 20. 21. 22. 23. 24. 25. 26. 27. 28. 29. 30. 31. 32. 33. 34. 35. 36. 37. 38. 39. 40. 41. 42. 43. 44. 45. 46. 47. 48. 49. 50. 51. 52. 53. 54. 55. 56. 57. 58. 59. 60. 61. 62. 63. 64. 65. 66. 67. 68. 69. 70. 71. 72. 73. 74. 75. 76. 77. 78. 79. 80. 81. 82. 83. 84. 85. 86. 87. 88. 89. 90. 91. 92. 93. 94. 95. 96. 97. 98. 99. 100.

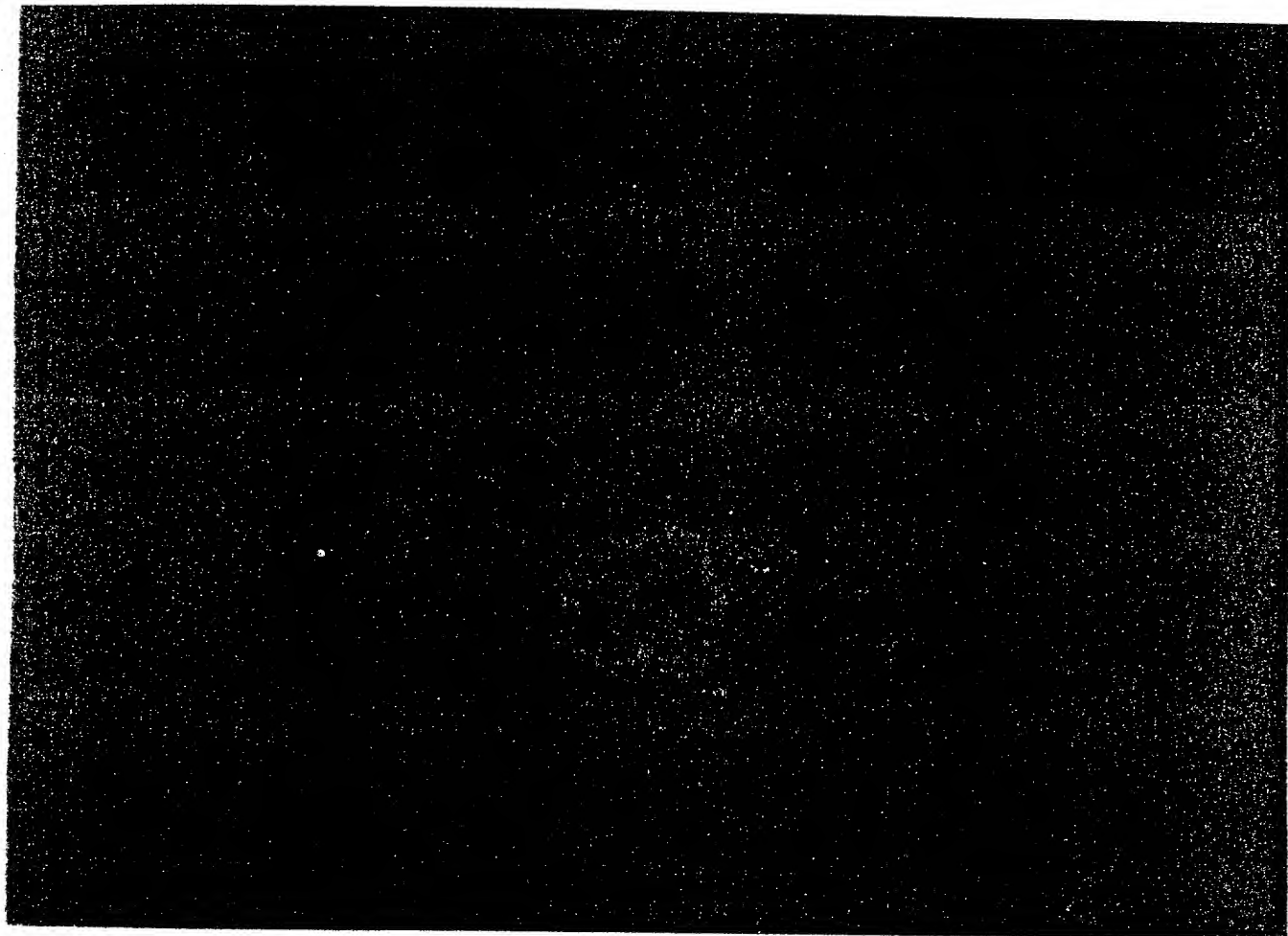


FIGURE 19

This is a high-contrast, black and white image, likely a scan of a book cover or endpaper. The image is characterized by a dense, grainy texture. A central, lighter, circular area is visible, surrounded by darker, more textured regions. The overall appearance is abstract and somewhat chaotic, with no discernible text or figures.

FIGURE 20

PSCA Immunostaining of Primary Tumors

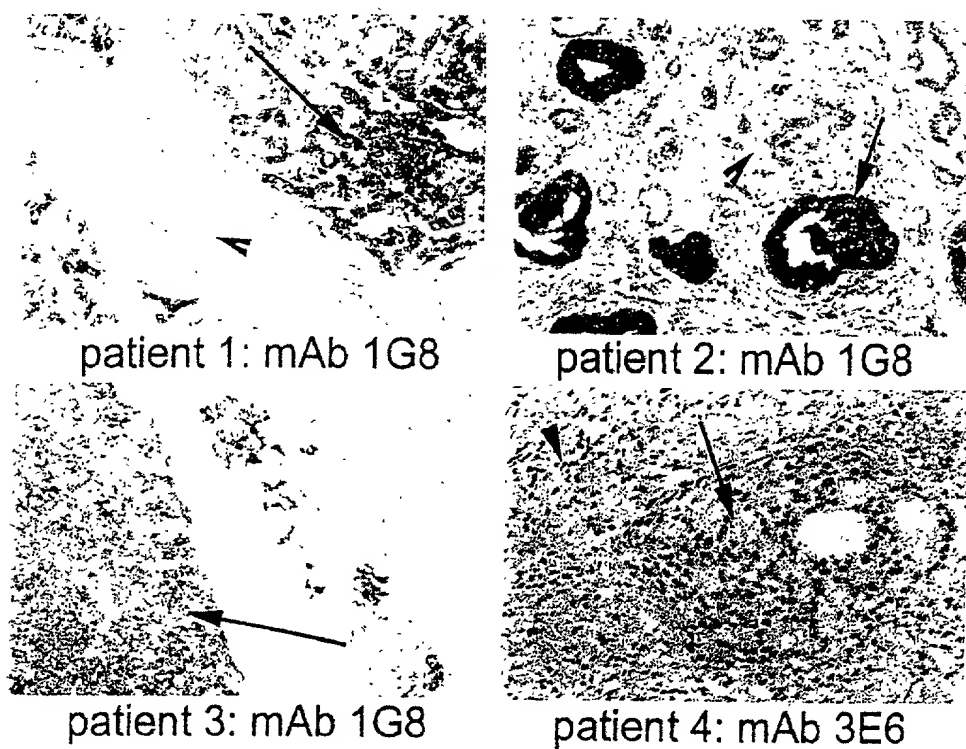


FIGURE 21

107.230" 2.24E660



FIGURE 22

401.330 624E660



FIGURE 23

FIGURE 24

[Faint handwritten notes at bottom left]

FIGURE 25

REF ID: A66666

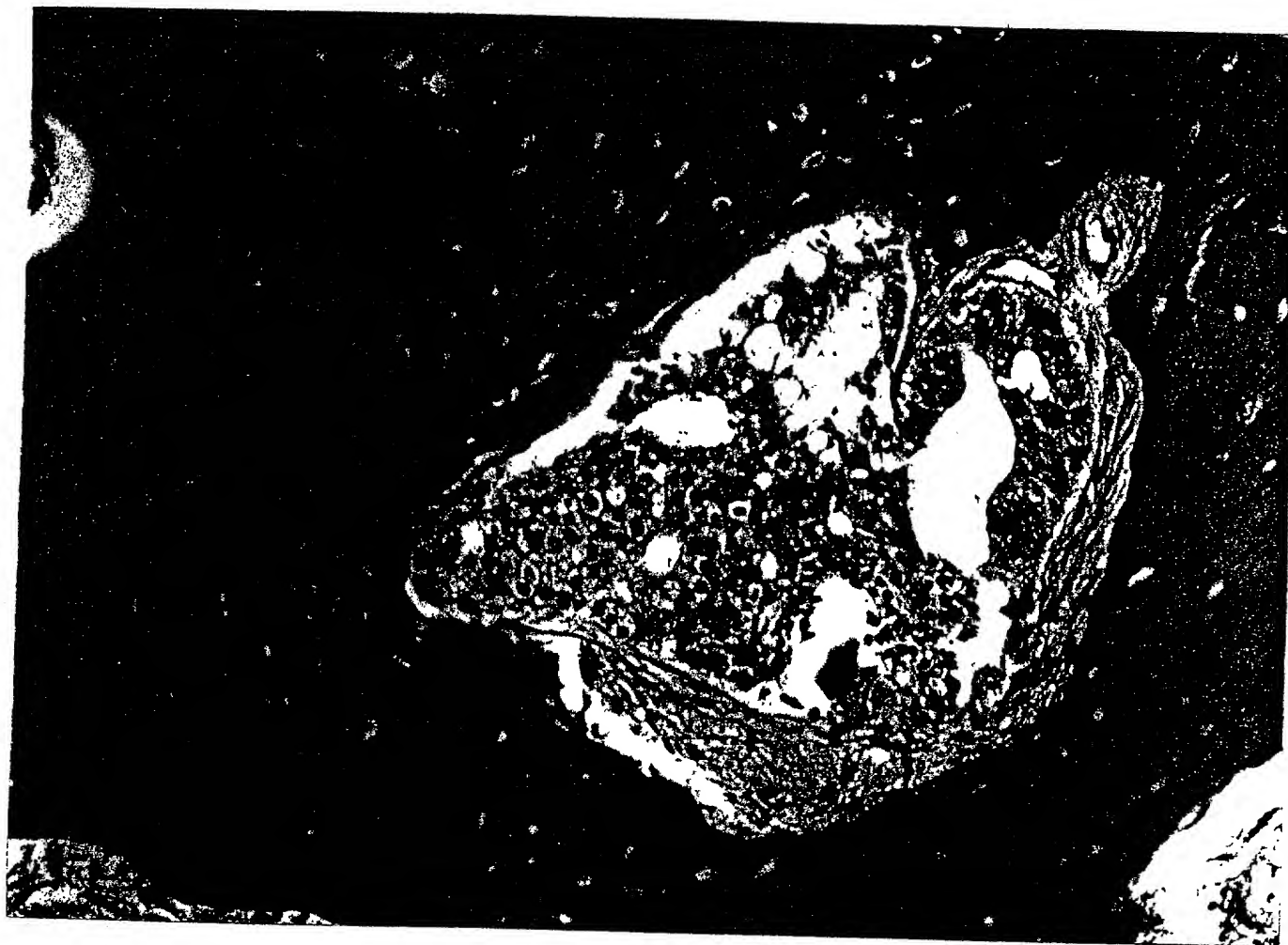
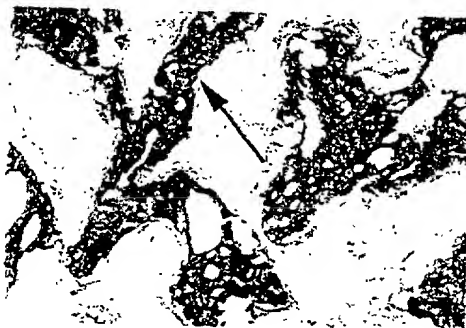


FIGURE 27

PSCA Immunostaining of Bony Metastases



Patient 5: H and E
and mAb 1G8



Patient 4: H and E
and mAb 3E6

FIGURE 28

094424650



FIGURE 29

107280 2/24/60

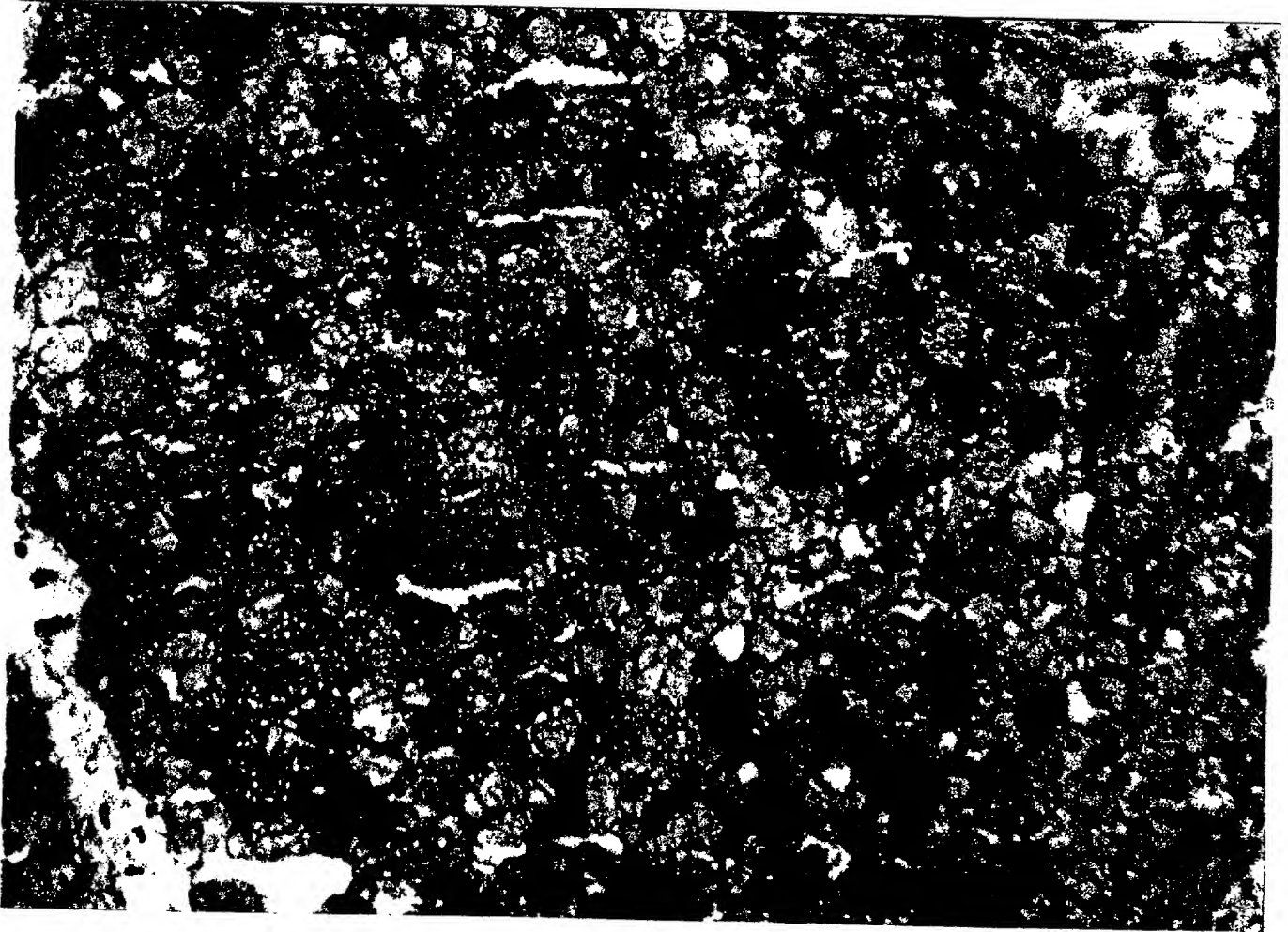


FIGURE 30

A high-contrast, black and white photograph showing a dense, textured surface, possibly a rock face or a wall. The image is characterized by a multitude of small, light-colored spots and patches, creating a speckled or mottled appearance against a dark background. The texture is irregular and granular, with some areas appearing more solid and others more eroded or pitted. The lighting is harsh, emphasizing the physical details of the surface.

FIGURE 31

This is a high-contrast, black and white image showing a dense, textured surface. The texture is mottled and irregular, with a mix of dark, almost black areas and lighter, greyish-white patches. The overall appearance is grainy and noisy, characteristic of a low-quality photocopy or a scan of a rough, aged surface like a book cover or endpaper. There are no discernible patterns, text, or figures.

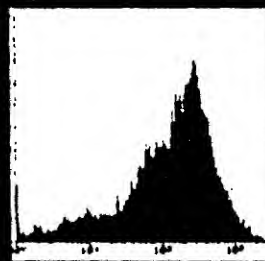
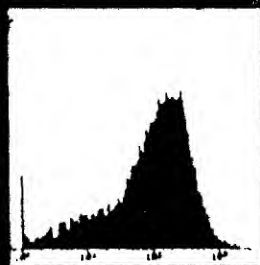
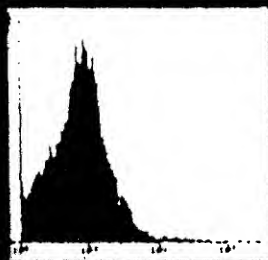
FIGURE 32

PSCA Expression in LAPC-9 Xenograft by FACS

Secondary Antibody

1G8

2H9



4A10

3C5

3E6

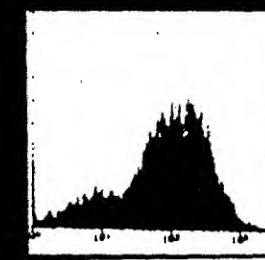
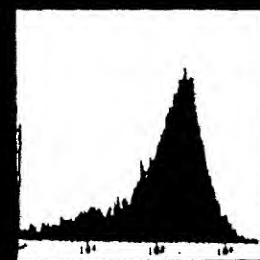
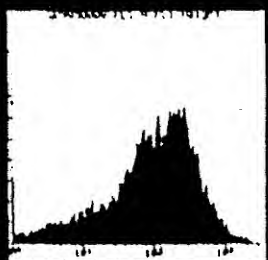


FIGURE 33

anti-PSCA mAbs

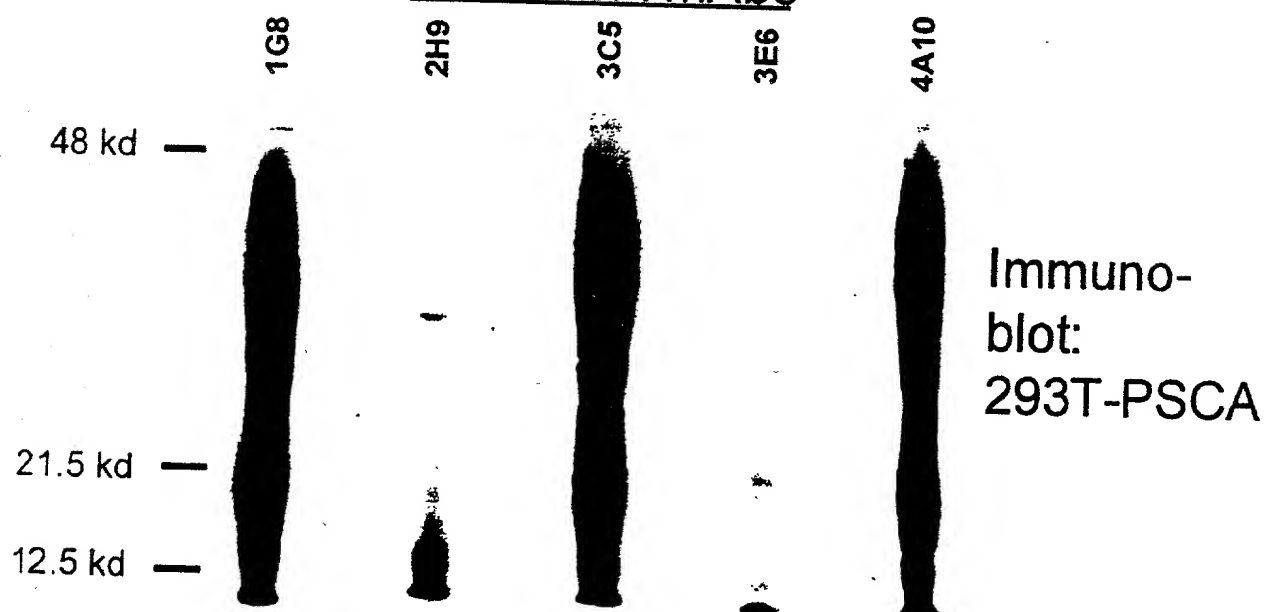


FIGURE 34

Immunofluorescent Staining of LNCaP-PSCA Cells

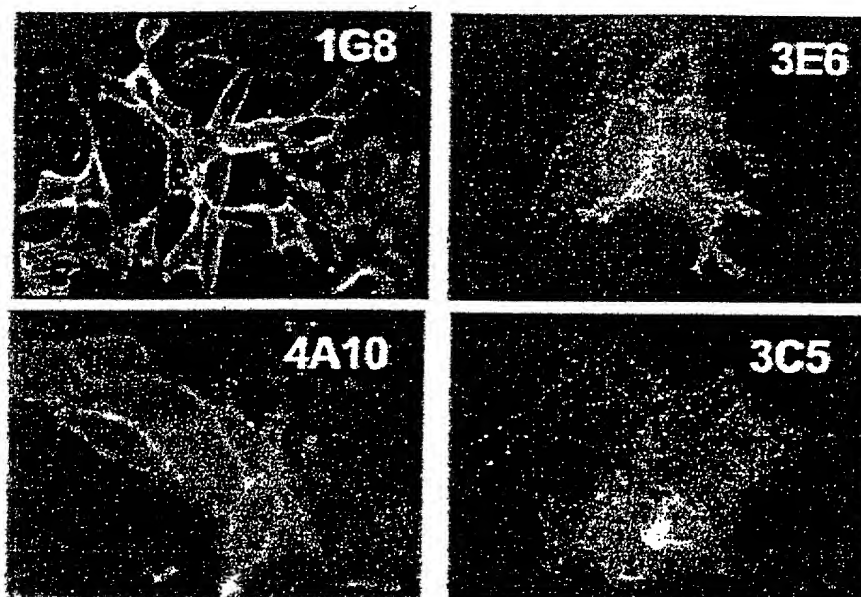


FIGURE 35

[illegible]

FIGURE 36

707 FEB 22 1990

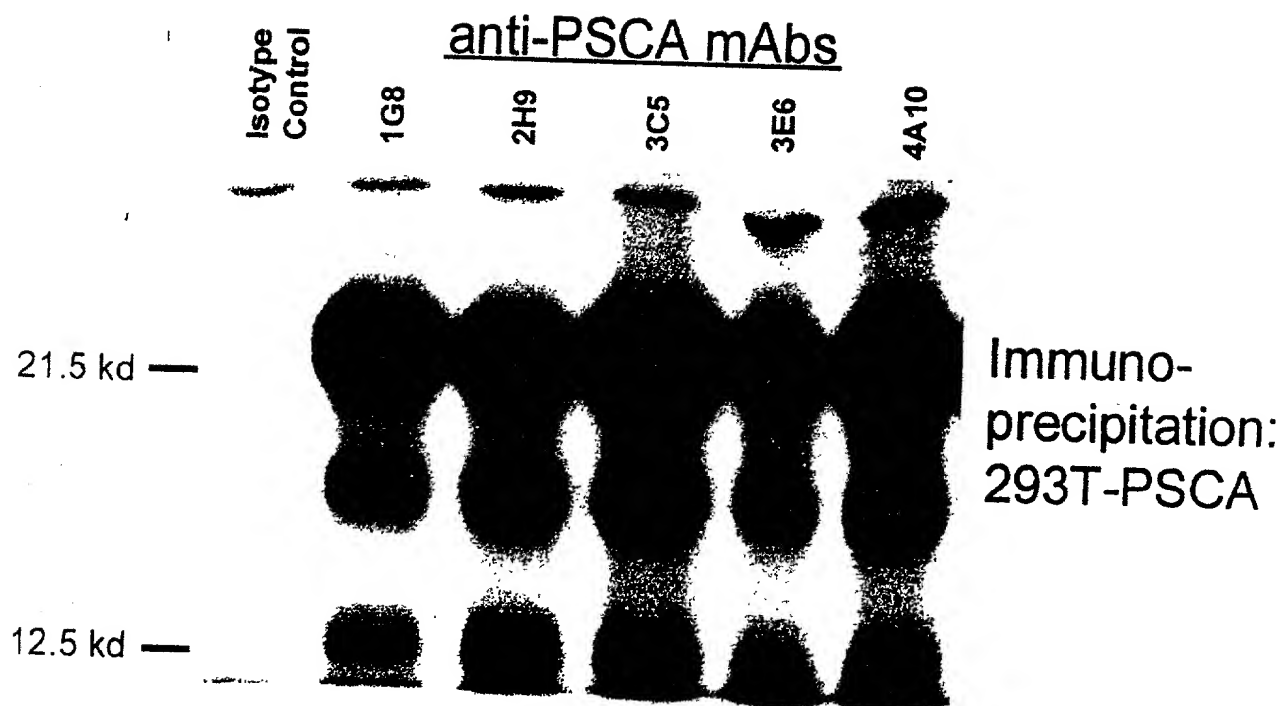


FIGURE 37

Immunohistochemical Staining of Normal Prostate

Normal: Isotype Control



Normal: PSCA mAb 3E6



Normal: PSCA mAb 1G8



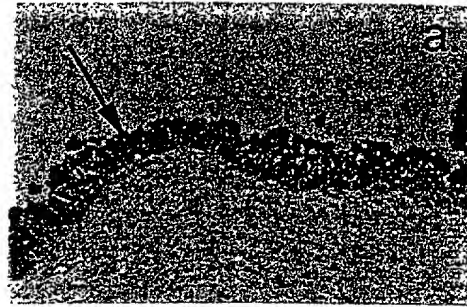
Atrophy: PSCA mAb 2H9



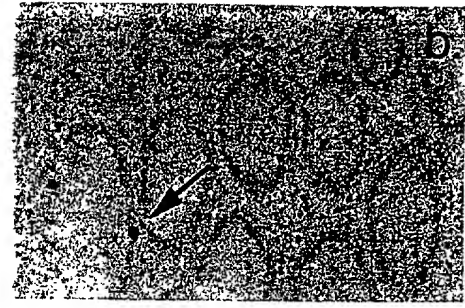
FIGURE 38

1.07293 E+06

A.



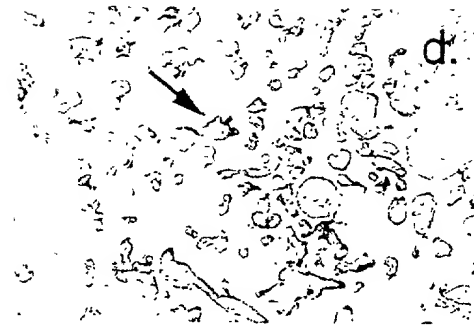
Bladder: 1G8



Colon: 1G8



Kidney: 3E6



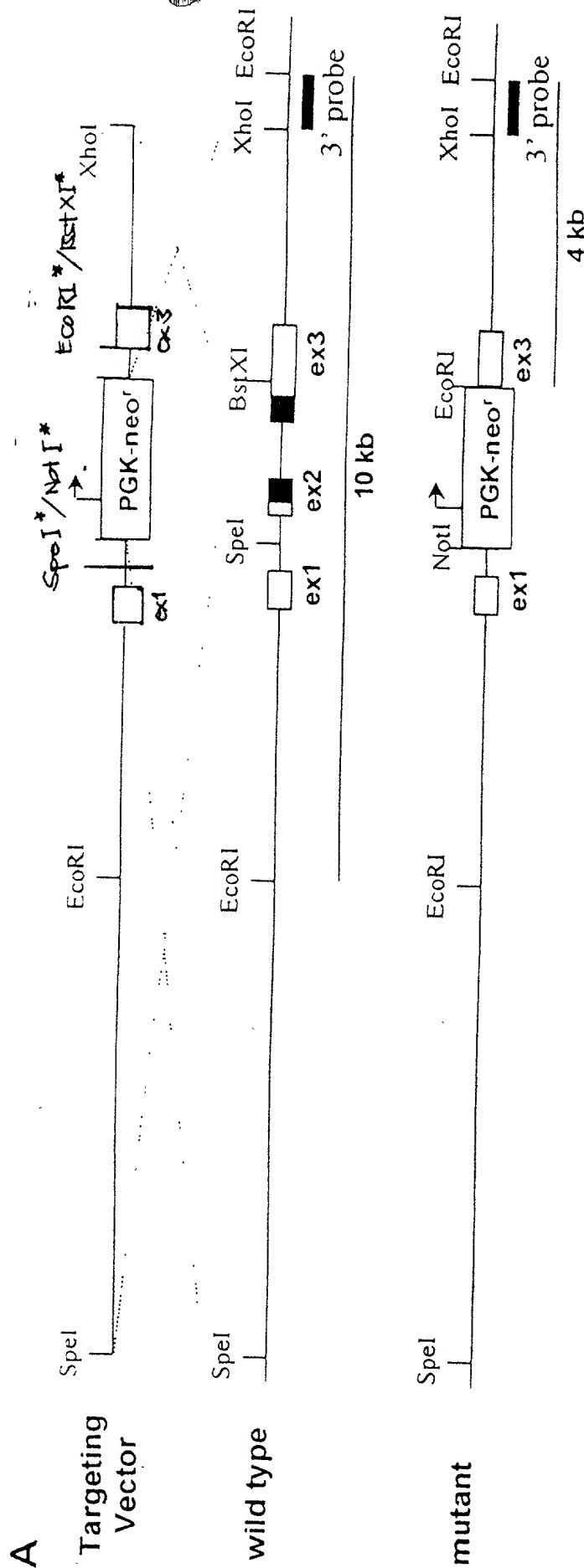
Placenta: 3E6

B.



FIGURE 39

Targeting of Mouse PSCA Gene



B. Genomic Southern Analysis of ES Cells

* ex1, 2, and 3 are the exons of PSCA gene.
 * Black boxes of ex2 and ex3 encode PSCA mature protein sequences.
 * ES genomic DNA's were digested with EcoRI, followed by Southern hybridization using 3' probe.

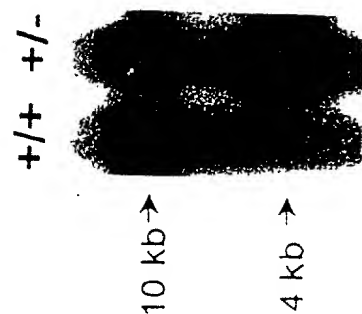
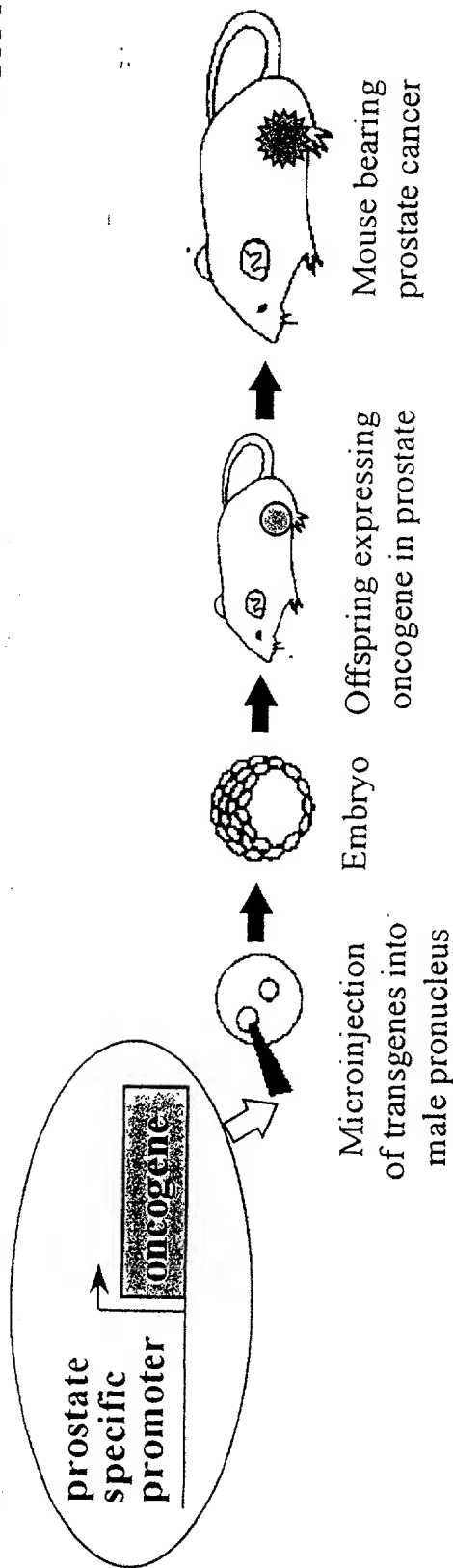


FIGURE 40

Transgenic Mouse Models of Prostate Cancer



| Transgene | Target tissues | Characteristics |
|--|--|---|
| C3(1) (-3 kb)/
SV40 large+small T
<i>Maroulakou et al.</i>
1994 <i>PNAS</i> | prostate (secretory cells)
urethral, mammary and
sweat gland | Low-grade PIN 8-12 wks
High-grade PIN 8-12 wks
Invasive carcinoma 28 wks
No metastases |
| Probasin (-426 bp)/
SV40 large+small T
<i>Greenberg et al.</i>
1995 <i>PNAS</i> | prostate (secretory cells) | Low-grade PIN 5-8 wks
High-grade PIN 8-12 wks
Invasive carcinoma 12 wks
Metastases in lymph node,
lung, liver and bone |
| Cryptdin2 (-6.5 kb)/
SV40 large+small T
<i>Garabedian et al.</i>
1998 <i>PNAS</i> | prostate
(neuroendocrine cells)
small intestine | Low-grade PIN 8-12 wks
High-grade PIN 8-12 wks
Invasive carcinoma 16 wks
Metastases in lymph node,
lung, liver and bone |

FIGURE 41

Reporter Gene Constructs for Transfection Assay

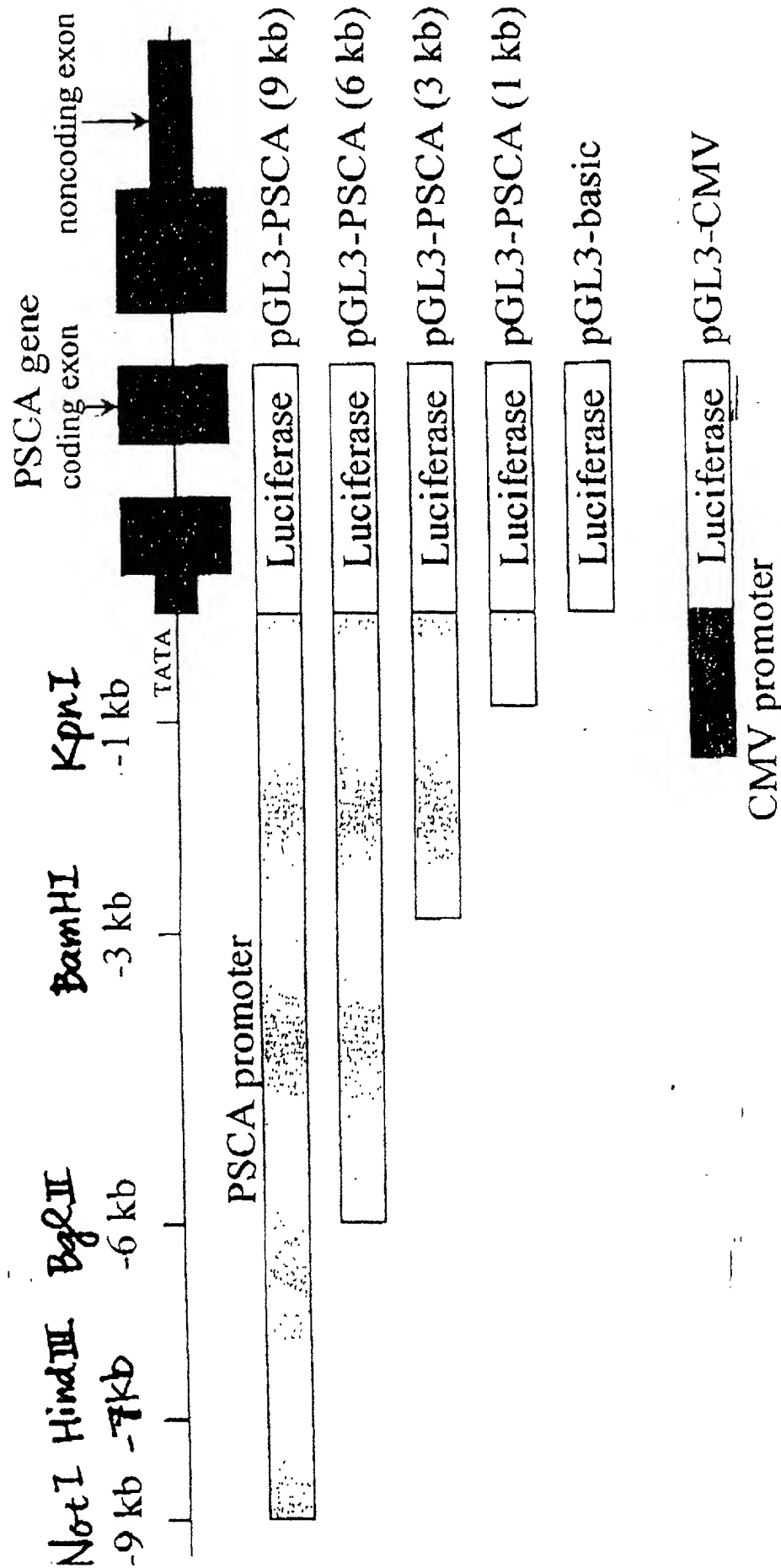


FIGURE 42

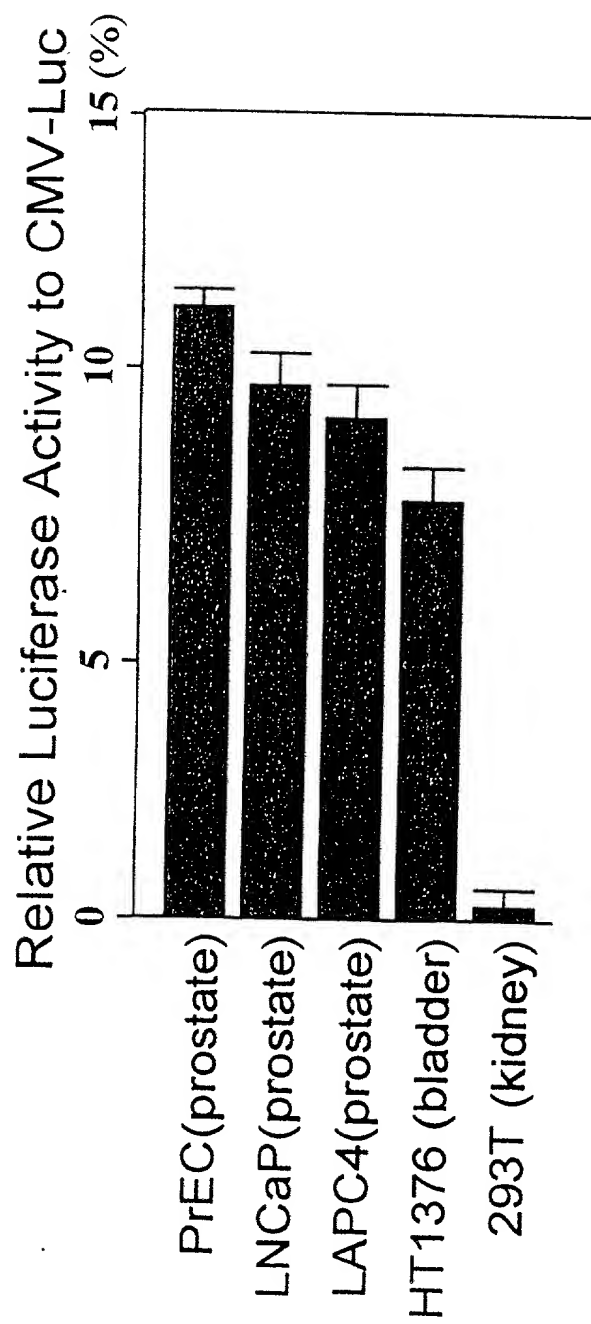


FIGURE 43

Identification of Prostate-Specific Elements Within PSCA Promoter Sequences

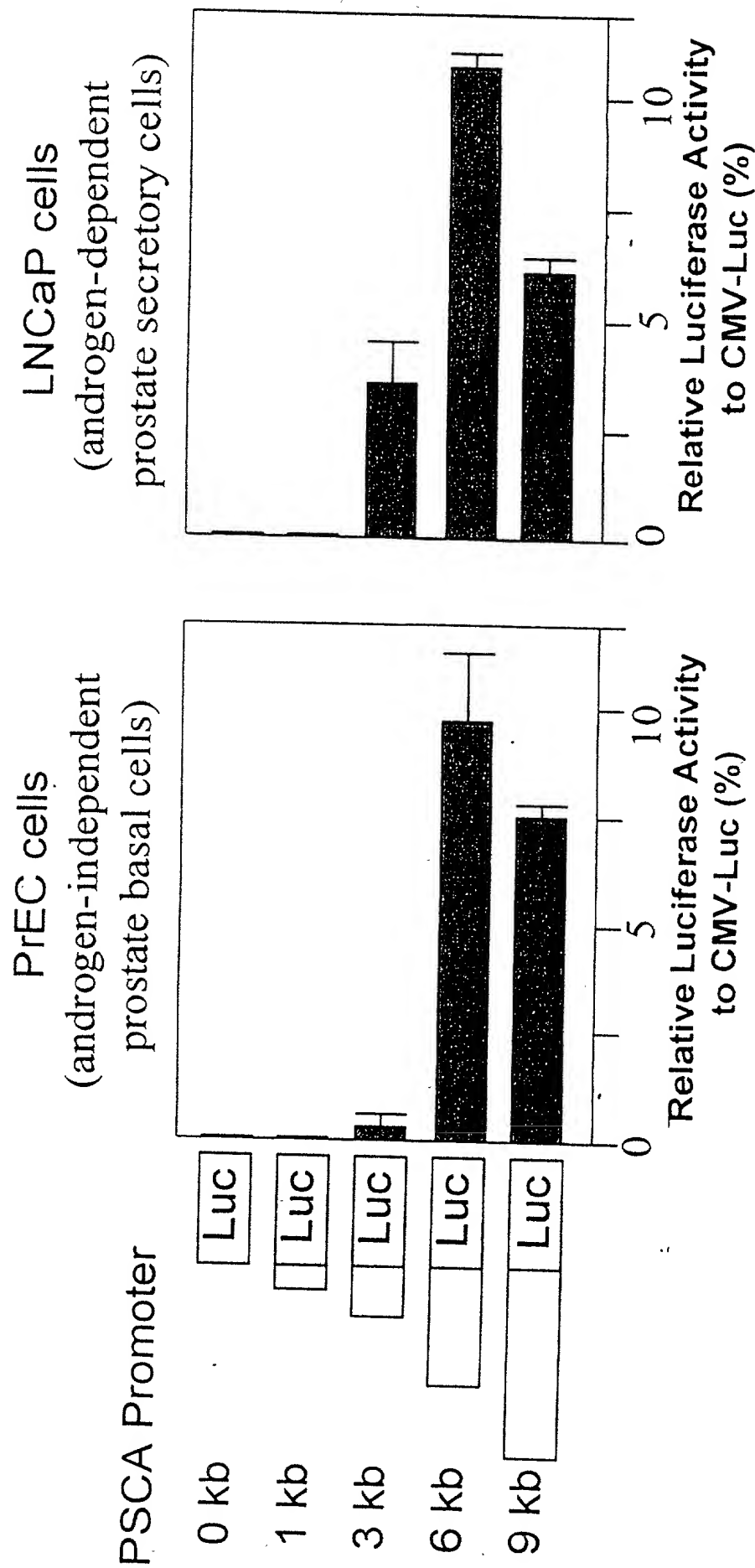


FIGURE 44

Update of Transgenic Mouse Projects

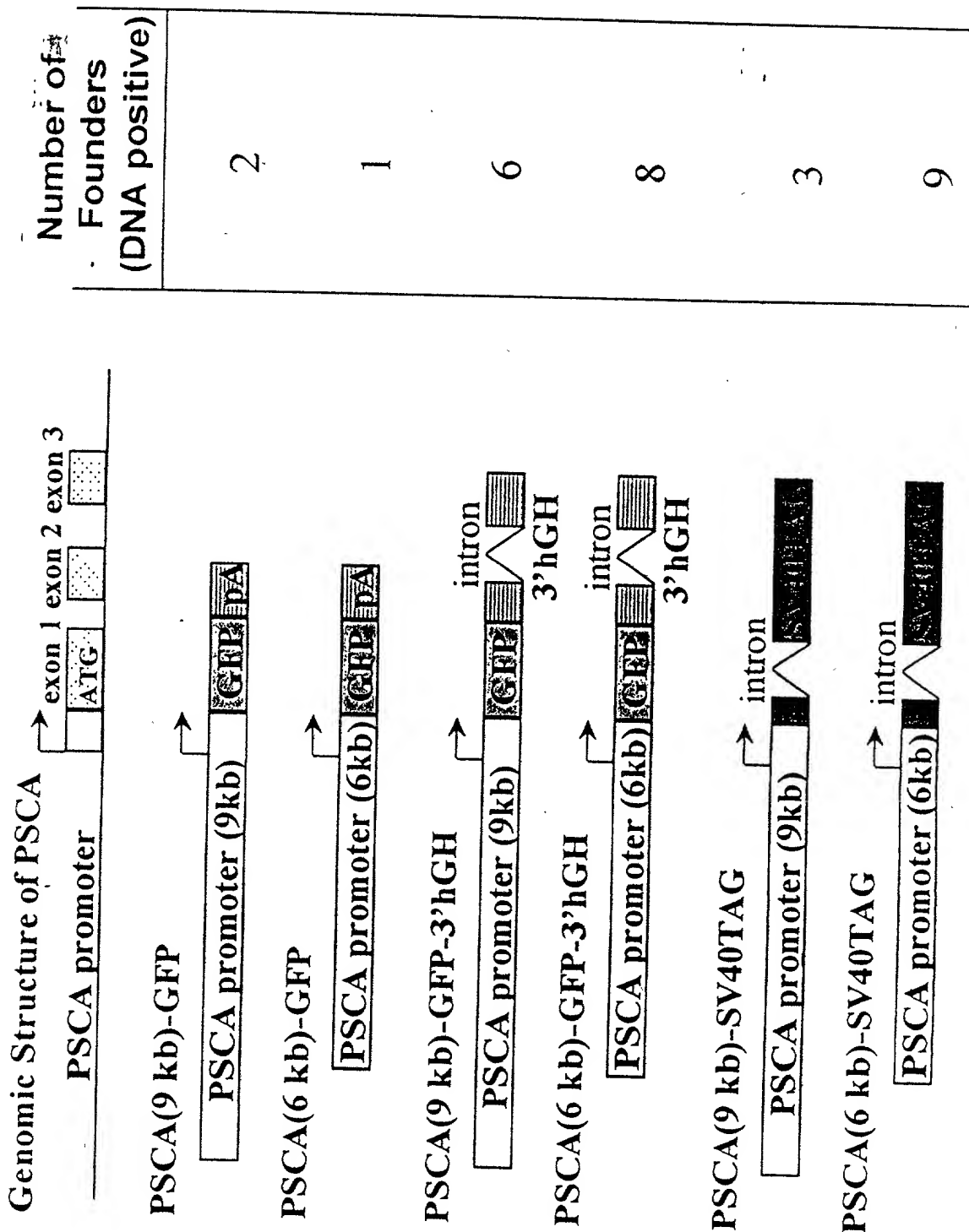
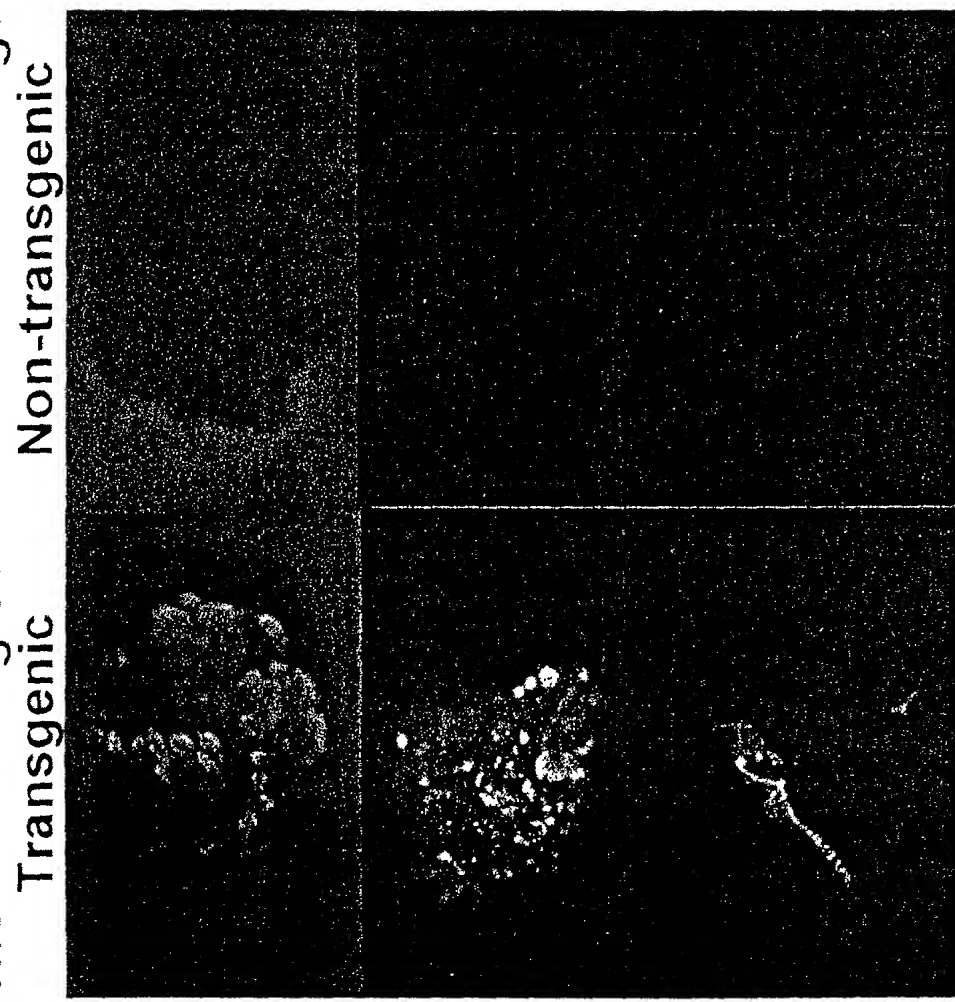


FIGURE 45

Whole-mount green fluorescence image



Negative tissues

- Stomach
- Small intestine
- Colon
- Seminal Vesicle
- Urethra
- Testis
- Liver
- Kidney
- Lung
- Brain
- Heart
- Skeletal muscle
- Ovary
- Uterus

Prostate
(A25-106-2)

Bladder
(A25-104)

Skin
(A25-106-2)

HUMAN
Spleen
Thymus
Prostate
Testis
Ovary
S. int.
Colon
PBL

Heart
Brain
Placenta
Lung
Liver
Muscle
Kidney
Panc.

hPSCA \rightarrow

Northern Analysis

MOUSE

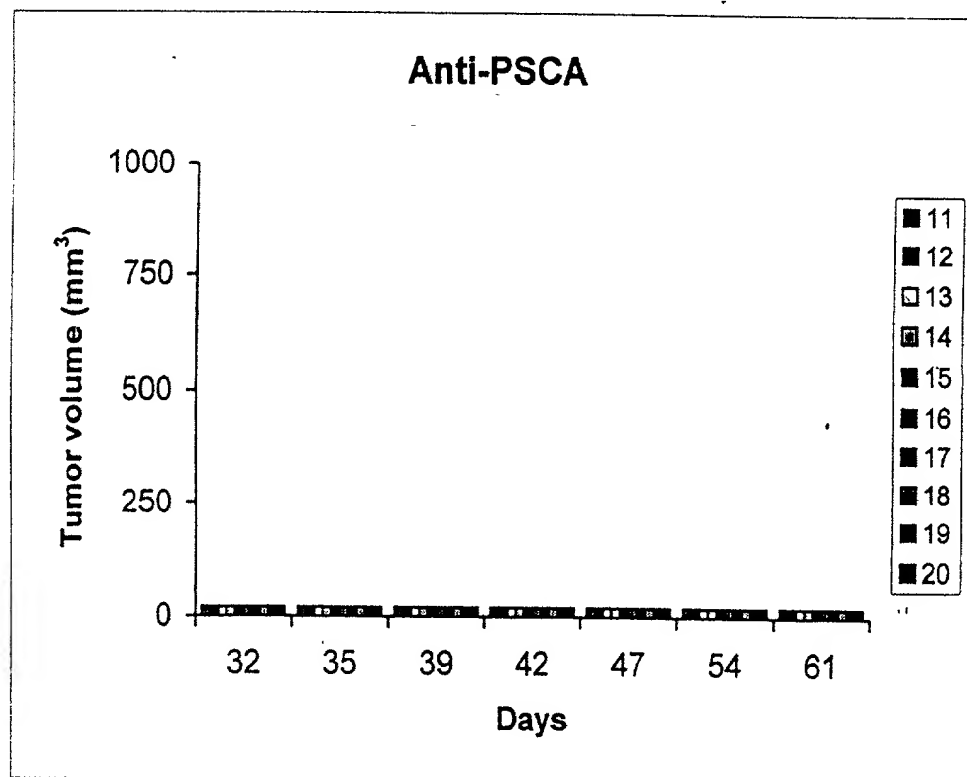
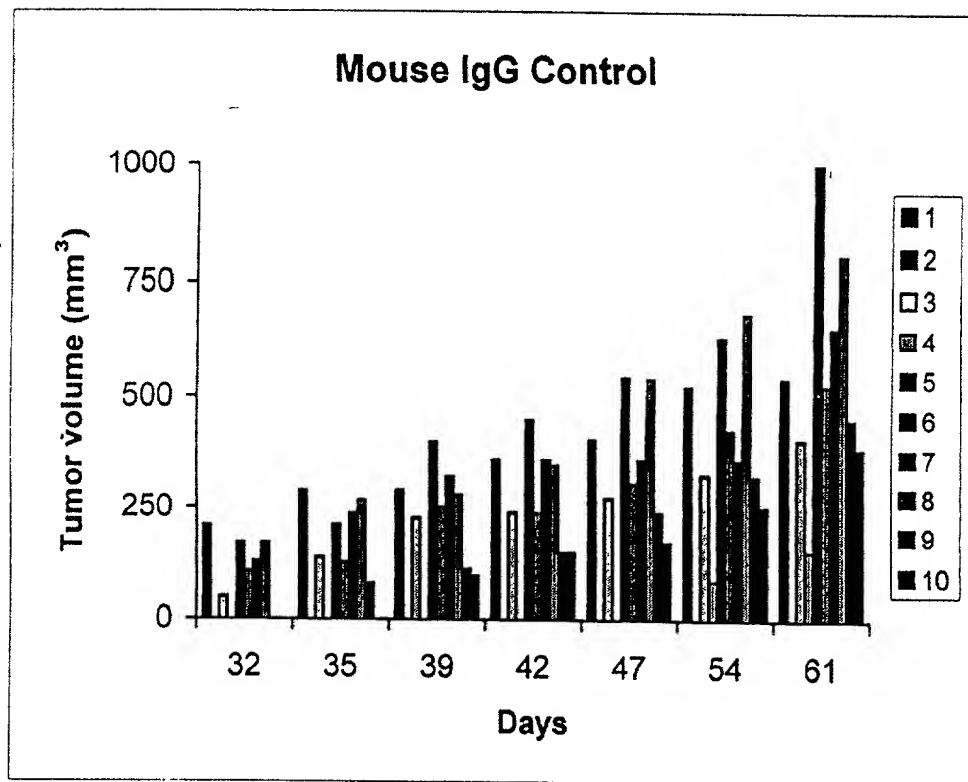
Ant. prostate
Dorso/Lat. prostate
Ventral prostate
Seminal vesicle
Urethral vesicle
Testis
Kidney

mPSCA \uparrow

mG3PDH \uparrow

RT-PCR

FIGURE 47



A

FIG. 49

Epitope recognized (OD 450 nm)

| <u>mAb</u> | <u>Isotype</u> | <u>F (18-98)</u> | <u>N (2-50)</u> | <u>M (46-109)</u> | <u>C (85-123)</u> |
|------------|----------------|------------------|-----------------|-------------------|-------------------|
| 1G8 | IgG1 k | 1.485 | 0.004 | 1.273 | 0.003 |
| 2A2 | IgG2a k | 0.973 | 0.631 | 0.023 | 0.010 |
| 2H9 | IgG1 k | 1.069 | 1.026 | 0.002 | 0.001 |
| 3C5 | IgG2a k | 1.916 | 1.709 | 0.006 | 0.002 |
| 3E6 | IgG3 k | 1.609 | 0.036 | 1.133 | 2.118 |
| 3G3 | IgG2a k | 2.805 | 1.731 | 0.004 | 0.000 |
| 4A10 | IgG2a k | 1.053 | 0.493 | 0.000 | 0.001 |

B

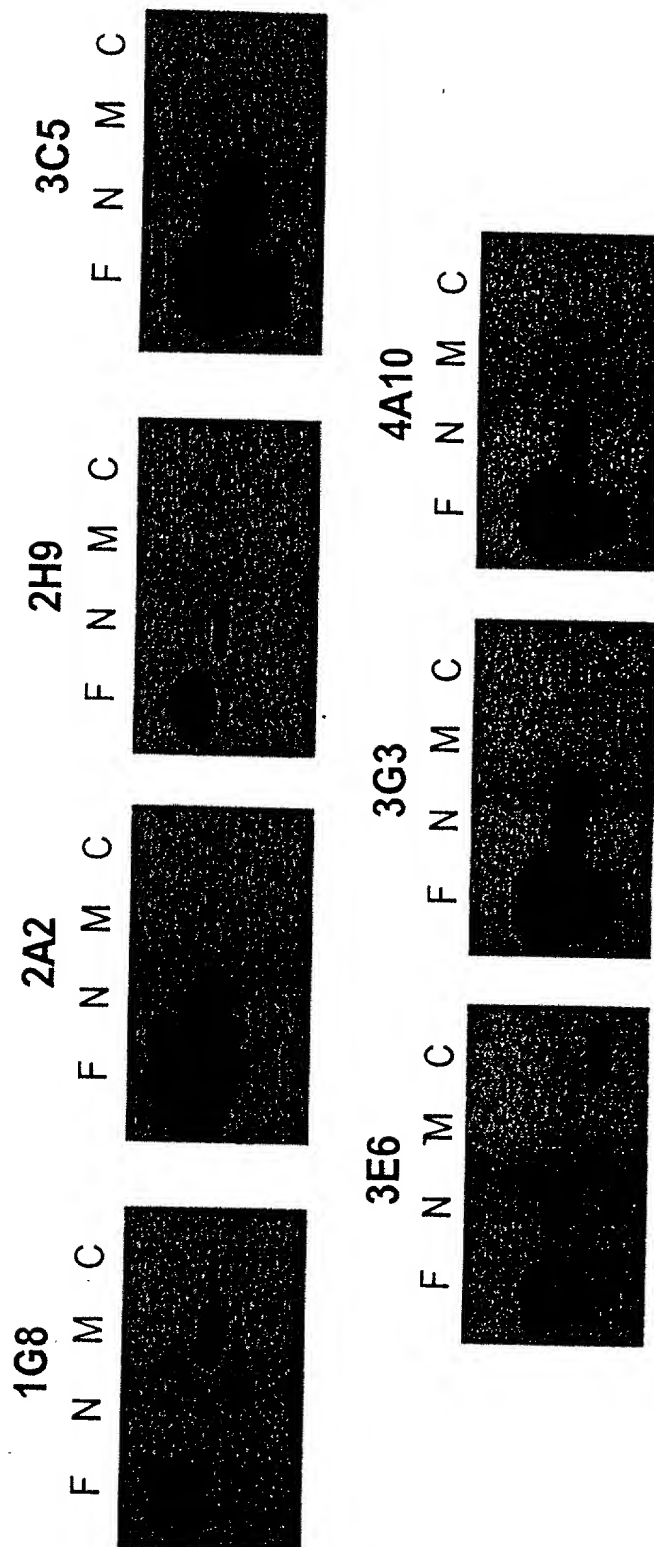
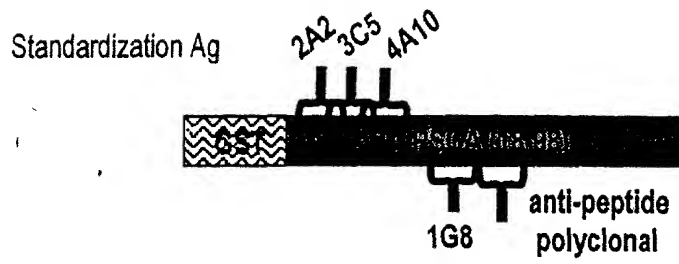
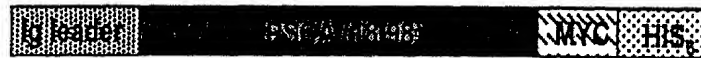


FIG. 50

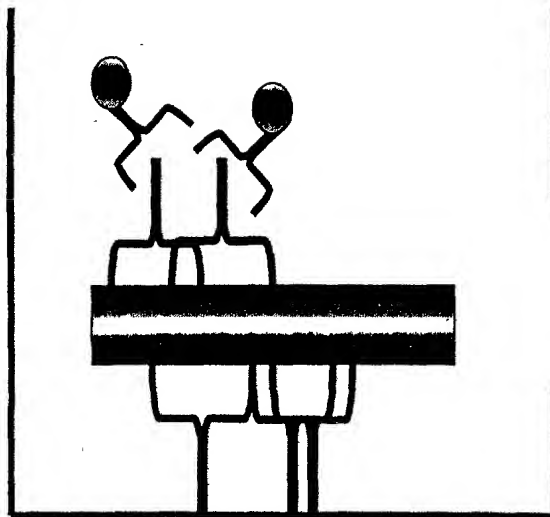
A



Engineered mammalian secreted form



B



Anti-IgG2a HRP

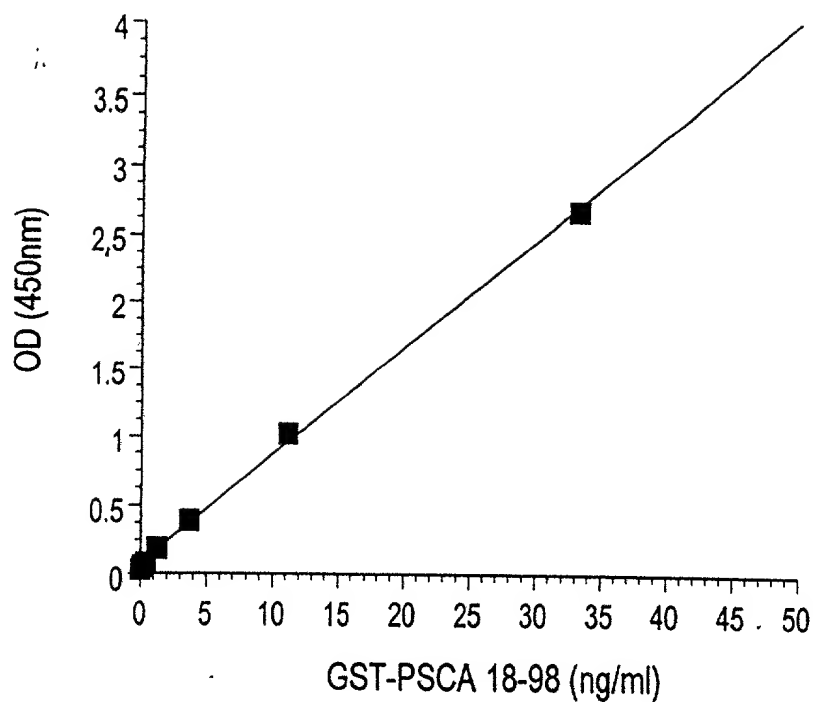
Anti-PSCA mAbs 3C5+4A10+2A2 (IgG2a)

PSCA

Affinity purified anti-peptide polyclonal
+ mAb 1G8 (IgG1)

FIG. 51

A



B

| <u>Sample</u> | <u>OD+range (n=2)</u> | <u>ng/ml</u> |
|------------------|-----------------------|--------------|
| vector | 0.005+0.001 | ND |
| vector+hu serum | 0.004+0.001 | ND |
| secPSCA | 2.695+0.031 | 32.92 |
| secPSCA+hu serum | 2.187+0.029 | 26.55 |

FIG. 52

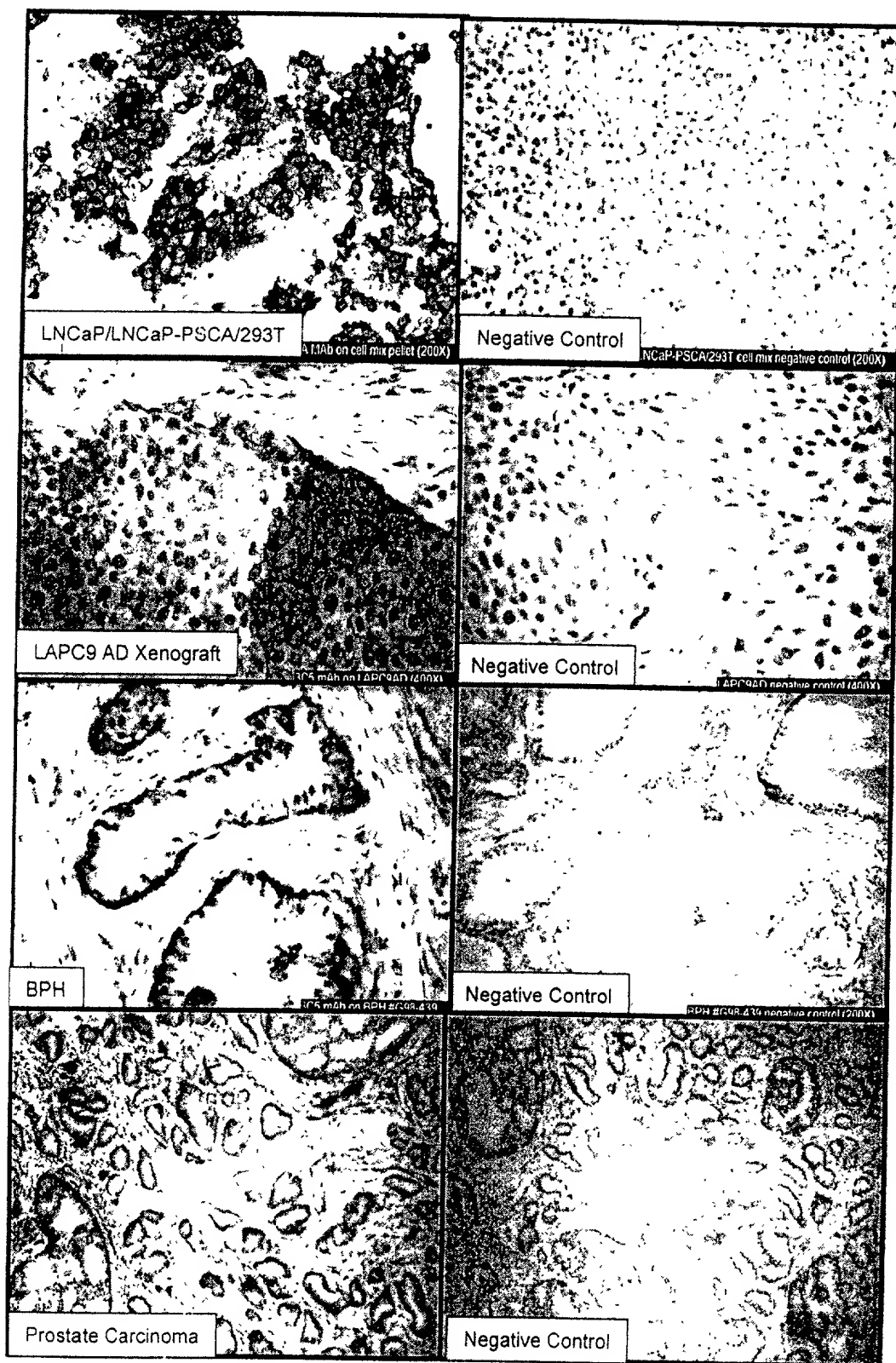


FIG. 53

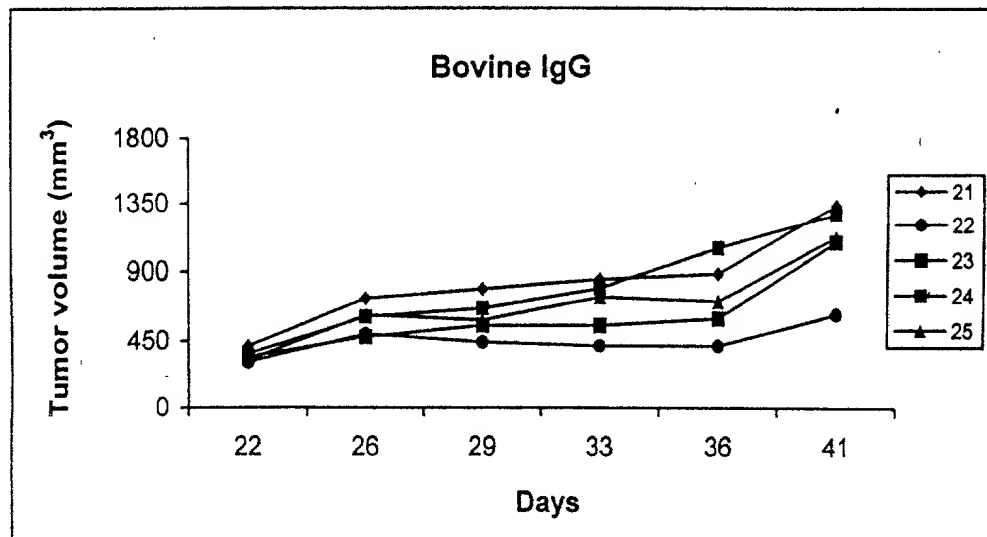
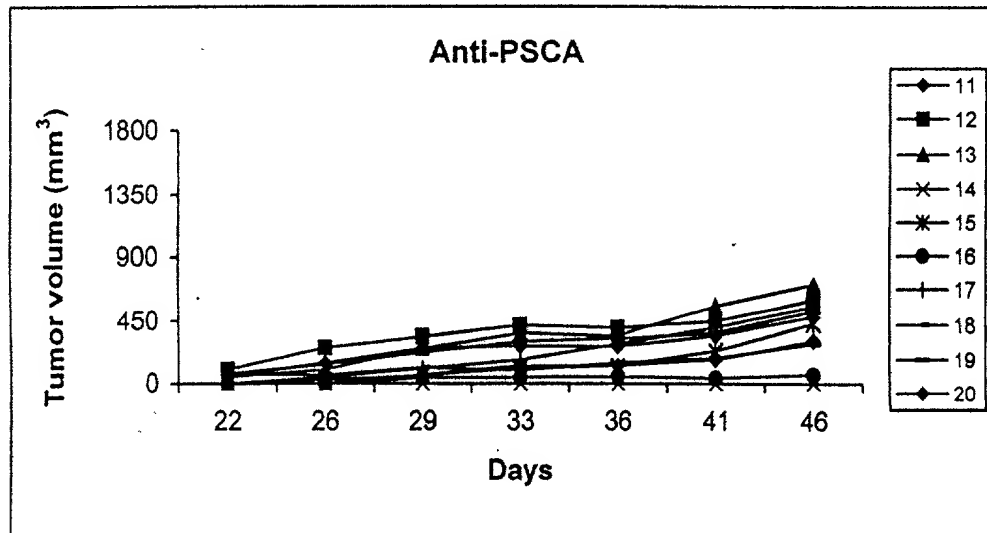
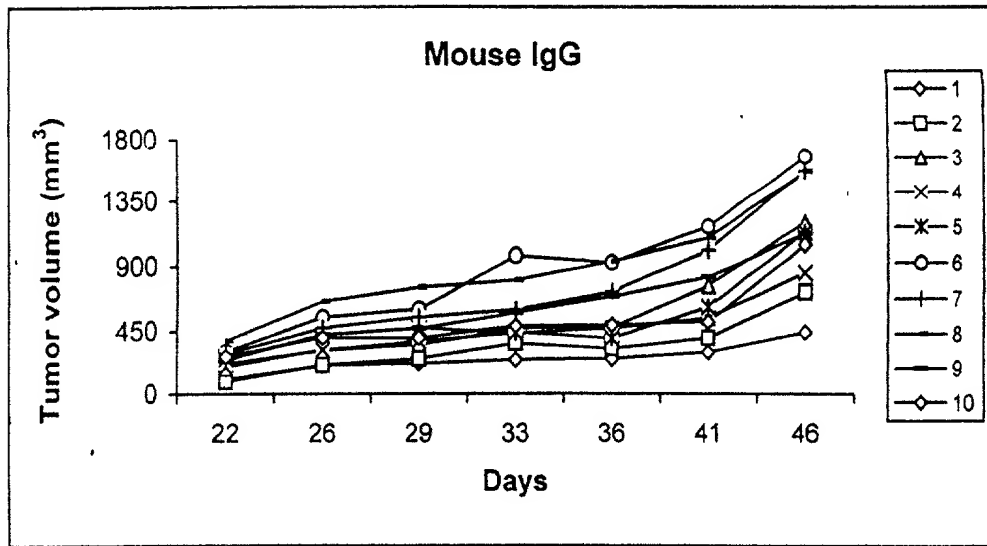


FIG. 54

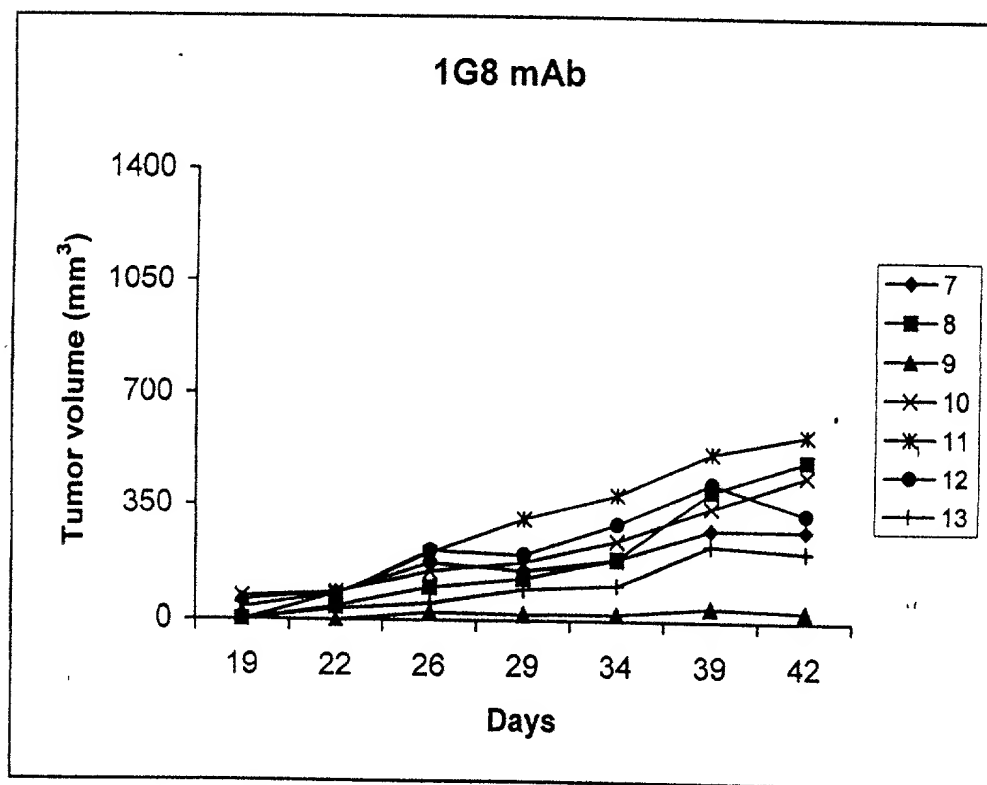
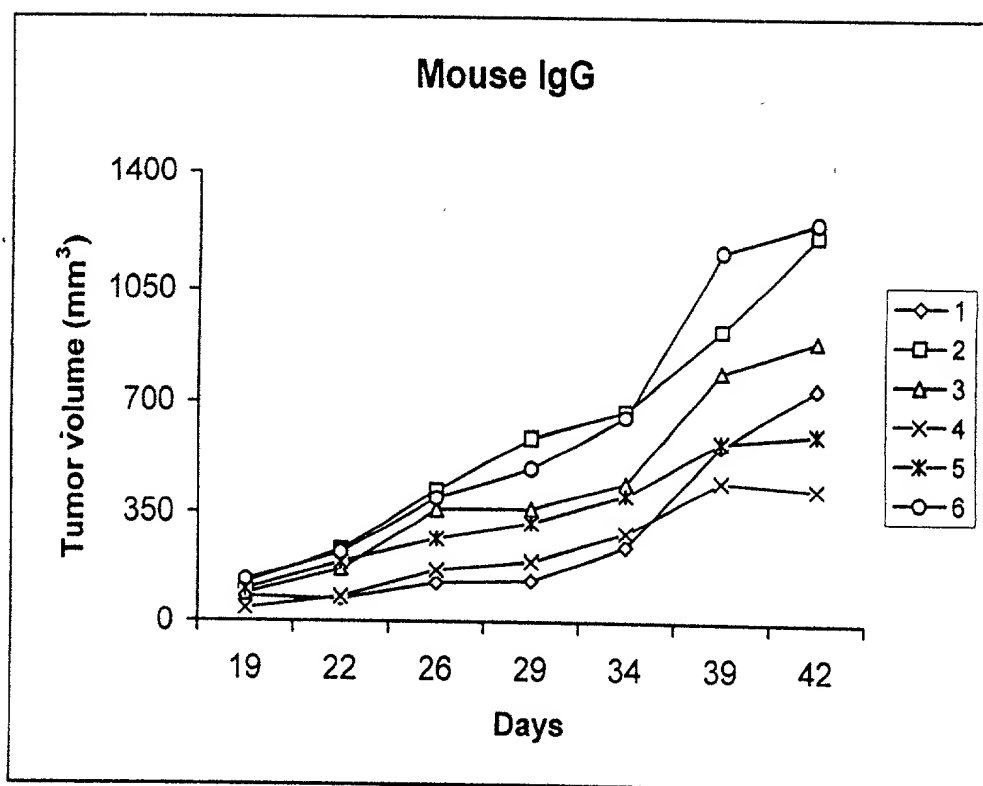


FIG. 55

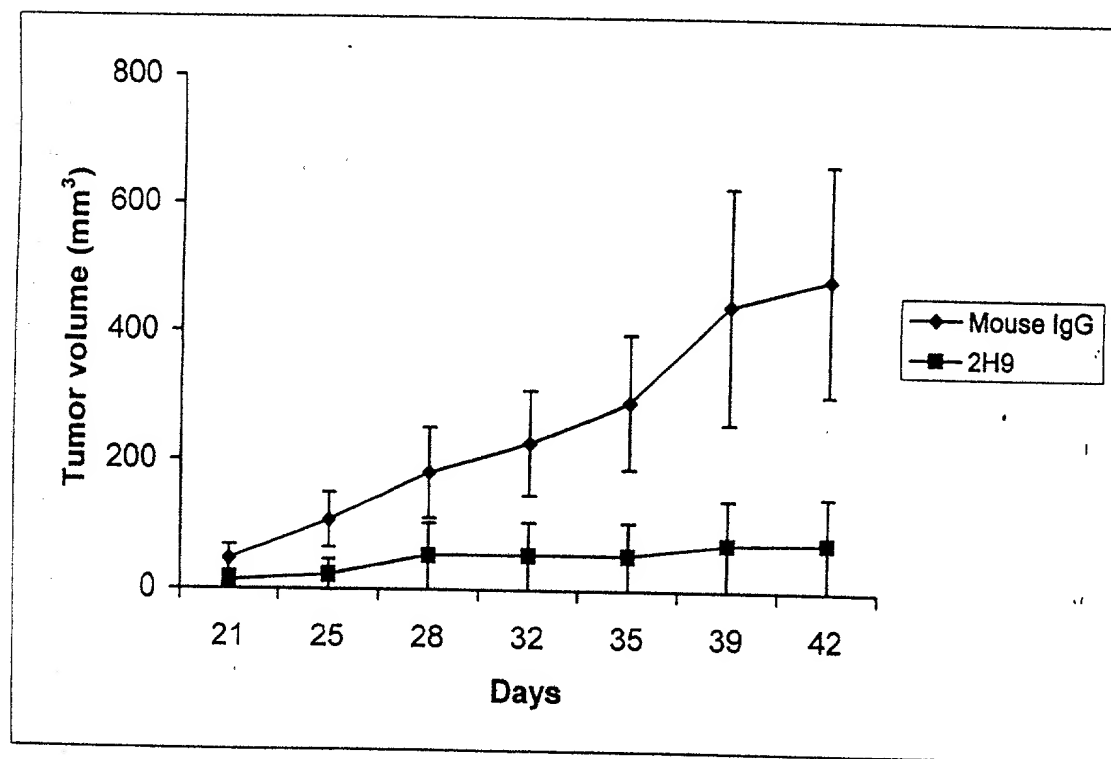
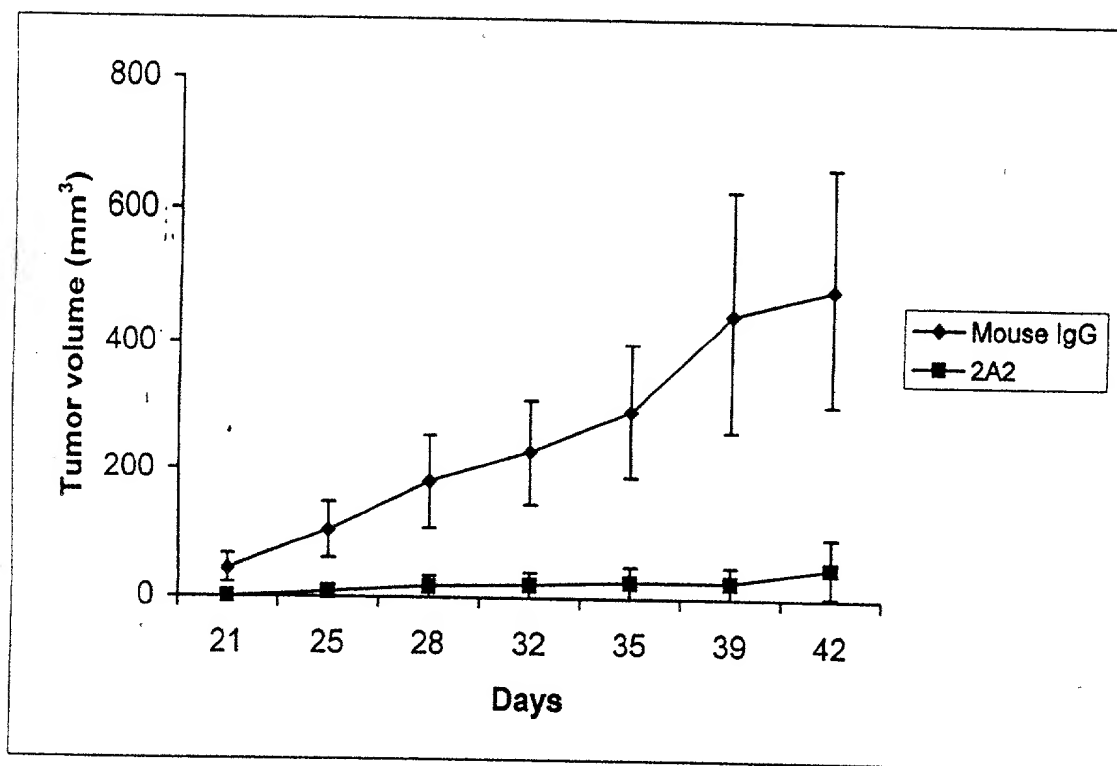


FIG. 56

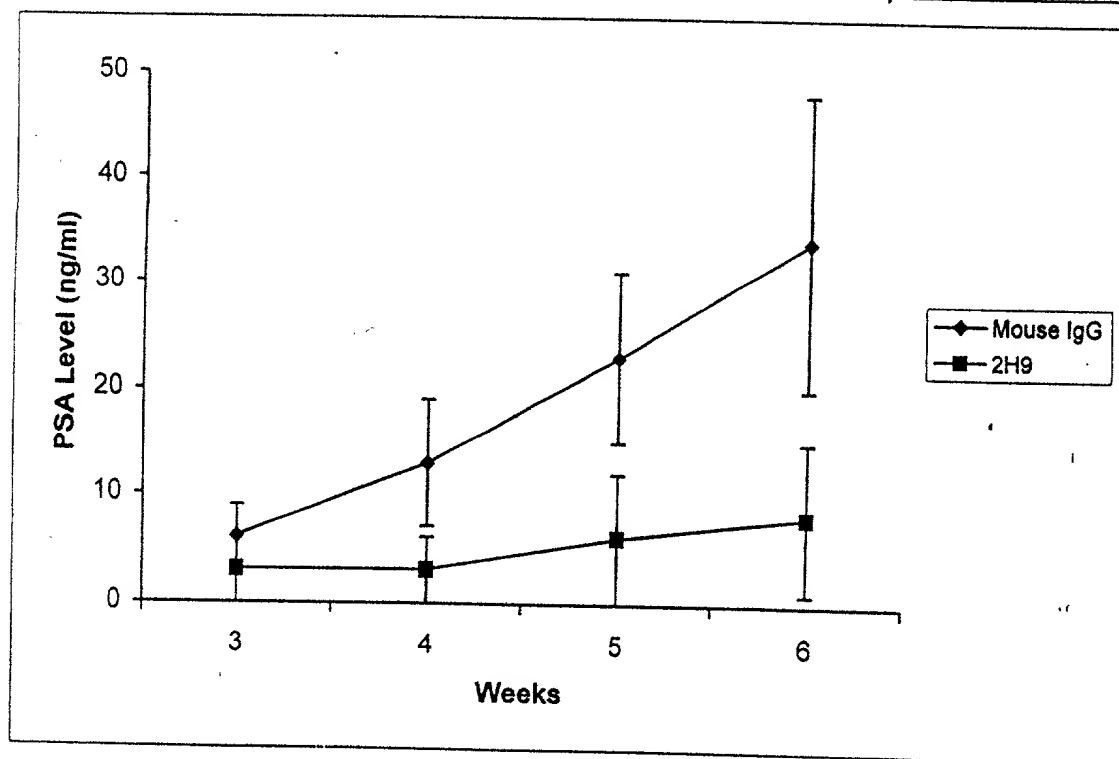
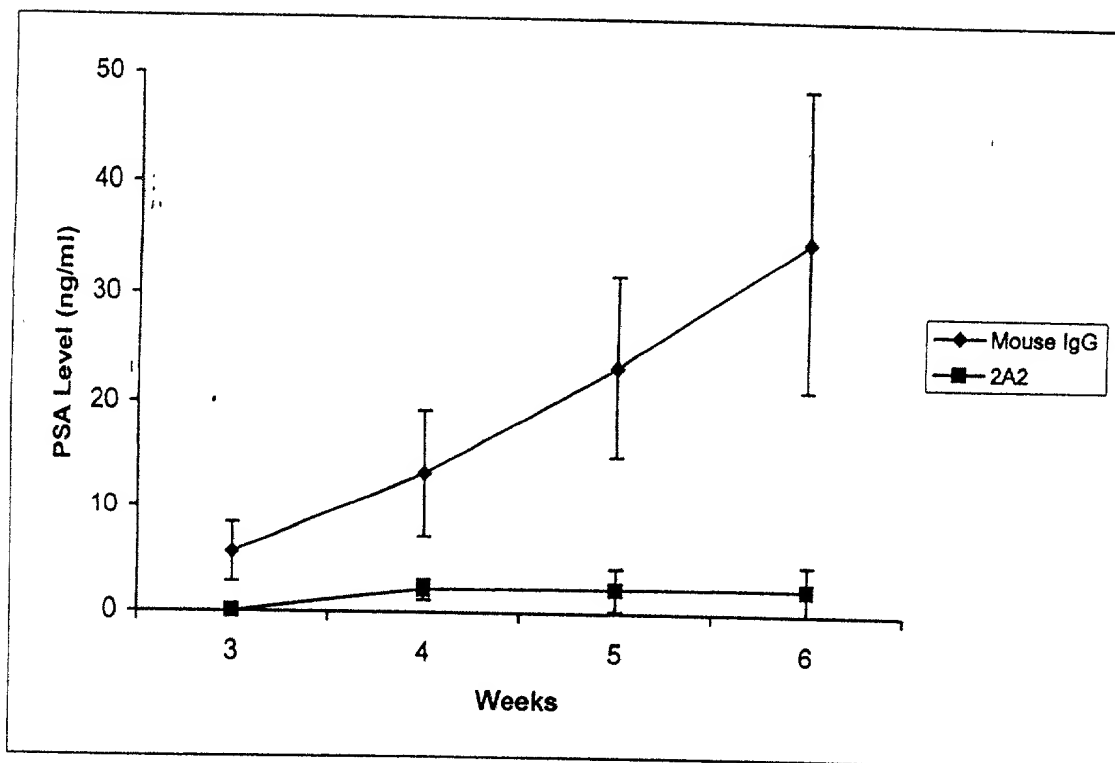


FIG. 57

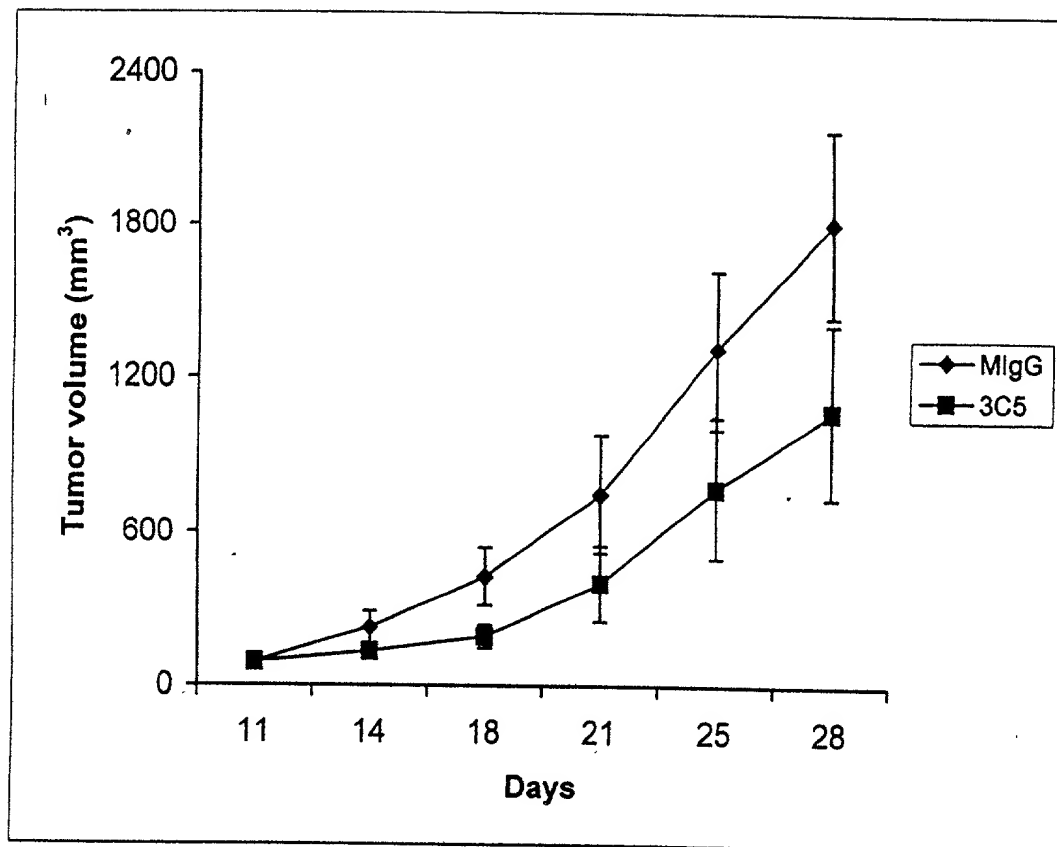


FIG. 58

TGCTTCTTCCTGATGGCAGTGGTTATAGGAGTCAATTCAGAGGTTTCAGCTGCAGCAGTCT 60
C F F L M A V V I G V N S E V Q L Q Q S 20

GGGGCAGAACTTGTGAGGTCAGGGGCCTCAGTCAAGTTGTCCTGCACAGCTTCTGGCTTC 120
G A E L V R S G A S V K L S C T A S G F 40

CDR1
AACATTAAAGACTACTATATACACTGGGTGAATCAGAGGCCTGACCAGGGCCTGGAGTGG 180
N I K D Y Y I H W V N Q R P D Q G L E W 60

CDR2
ATTGGATGGATTGATCCTGAGAATGGTGACACTGAATTTGTCCCGAAGTTCCAGGGCAAG 240
I G W I D P E N G D T E F V P K F Q G K 80

GCCACTATGACTGCAGACATTTTCTCCAACACAGCCTACCTGCACCTCAGCAGCCTGACA 300
A T M T A D I F S N T A Y L H L S S L T 100

CDR3
TCTGAAGACACTGCCGTCTATTACTGTAAAACGGGGGGTTTCTGGGGCCAAGGGACTCTG 360
S E D T A V Y Y C K T G G F W G Q G T L 120

GTCACTGTCTCTGCAGCCAAAACGACACCCCCATCTGTCTATCCACTG
V T V S A A K T T P P S V Y P L

FIG. 58

FIG. 59

TTGGTAGCAACAGCCTCAGATGTCCACTCCCAGGTCCAAGTGCAGCAACCTGGGTCTGAA 60
L V A T A S D V H S Q V Q L Q Q P G S E 20

CTGGTGAGGCCTGGAAGTTCAGTGAAGCTGTCCTGCAAGGCTTCTGGCTATACATTCTCC 120
L V R P G T S V K L S C K A S G Y T F S 40
CDR1

AGCTACTGGATGCACTGGGTGAAGCAGAGGCCTGGACAAGGCCTTGAGTGGATTGGAAAT 180
S Y W M H W V K Q R P G Q G L E W I G N 60

ATTGACCCTGGTAGTGGTTACACTAACTACGCTGAGAACCTCAAGACCAAGGCCACACTG 240
I D P G S G Y T N Y A E N L K T K A T L 80
CDR2

ACTGTAGACACATCCTCCAGCACAGCCTACATGCAGCTCAGCAGCCTGACATCTGAGGAC 300
T V D T S S S T A Y M Q L S S L T S E D 100

TCTGCAGTCTATTACTGTACAAGCCGATCTACTATGATTACGACGGGATTGCTTACTGG 360
S A V Y Y C T S R S T M I T T G F A Y W 120
CDR3

GGCCAAGGGACTCTGGTCACTGTCTCTGCAGCTACAACAACAGCCCCATCTGTCTATCCA 420
G Q G T L V T V S A A T T T A P S V Y P 160

CTGGCC
L A

FIG. 59

FIG. 60

AATGACTTCGGGTTGAGCTGGGTTTTTATTATTGTTCTTTTAAAAGGGGTCCGGAGTGAA 60
N D F G L S W V F I I V L L K G V R S E 20

GTGAGGCTTGAGGAGTCTGGAGGAGGCTGGGTGCAACCTGGAGGATCCATGAAACTCTCC 120
V R L E E S G G G W V Q P G G S M K L S 40

TGTGTAGCCTCTGGATTTACTTTTCAGTAATTACTGGATGACTTGGGTCCGCCAGTCTCCA 180
C V A S G F T F S N Y W M T W V R Q S P 60
CDR1

GAGAAGGGGCTTGAGTGGGTTGCTGAAATTCGATTGAGATCTGAAAATTATGCAACACAT 240
E K G L E W V A E I R L R S E N Y A T H 80
CDR2

TATGCGGAGTCTGTGAAAGGGAAATTCACCATCTCAAGAGATGATTCCAGAAGTCGTCTC 300
Y A E S V K G K F T I S R D D S R S R L 100

TACCTGCAAATGAACAACTTAAGACCTGAAGACAGTGGAATTTATTACTGTACAGATGGT 360
Y L Q M N N L R P E D S G I Y Y C T D G 120

CTGGGACGACCTAACTGGGGCCAAGGGACTCTGGTCACTGTCTCTGCAGCCAAAACGACA 420
L G R P N W G Q G T L V T V S A A K T T 140
CDR3

CCCCCATCTGTCTATCCACTGGCCCCTTGTGTA
P P S V Y P L A P C V

107259 E24660

FIG. 63

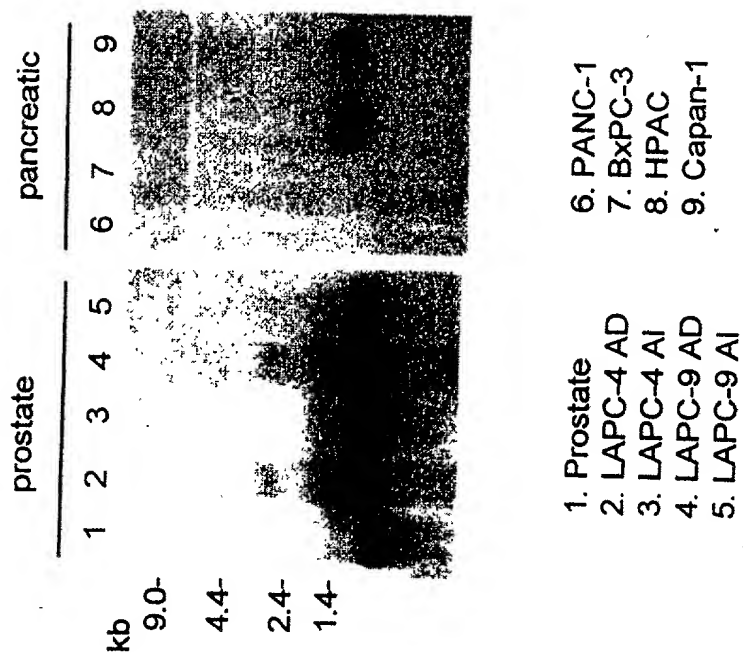
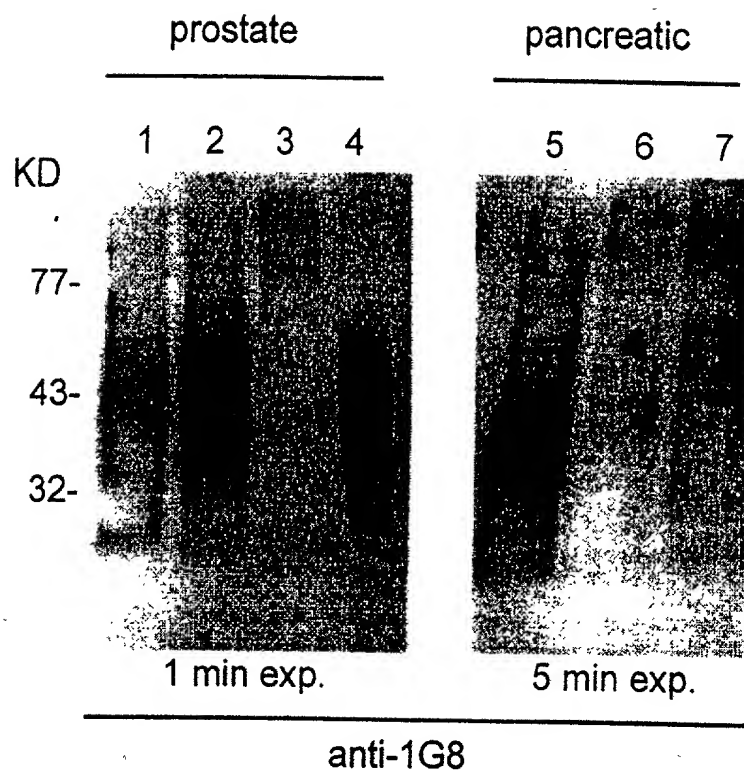


FIG. 64



1. LAPC-4 AD
2. LAPC-9 AI
3. LNCaP
4. LNCaP-PSCA

5. HPAC
6. Capan-1
7. ASPC-1

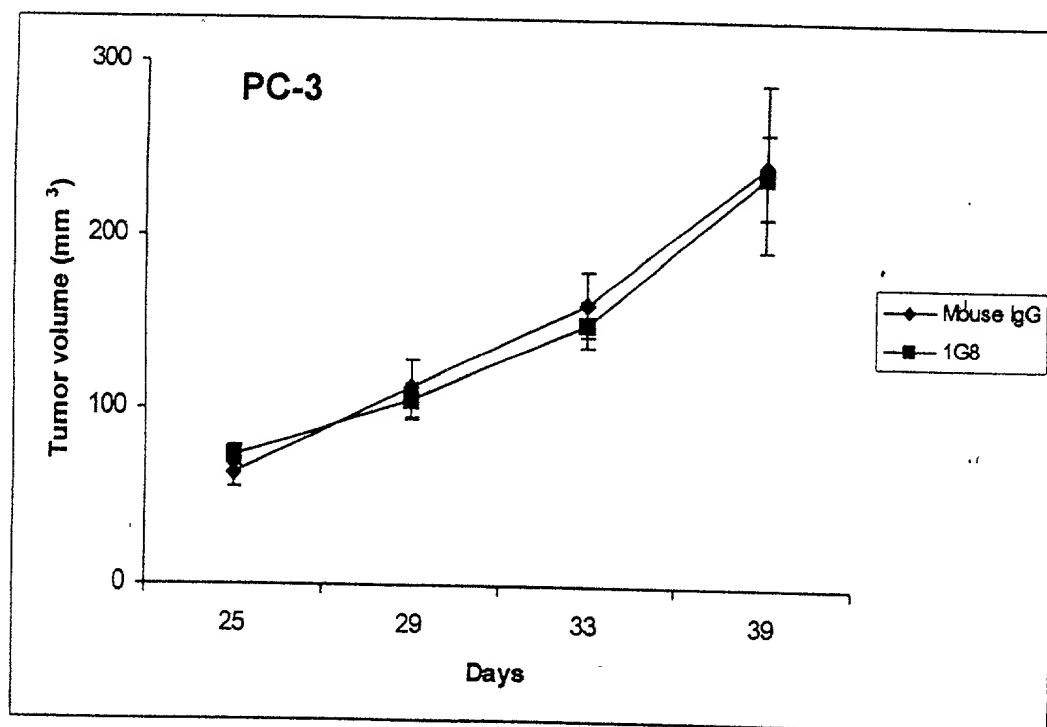
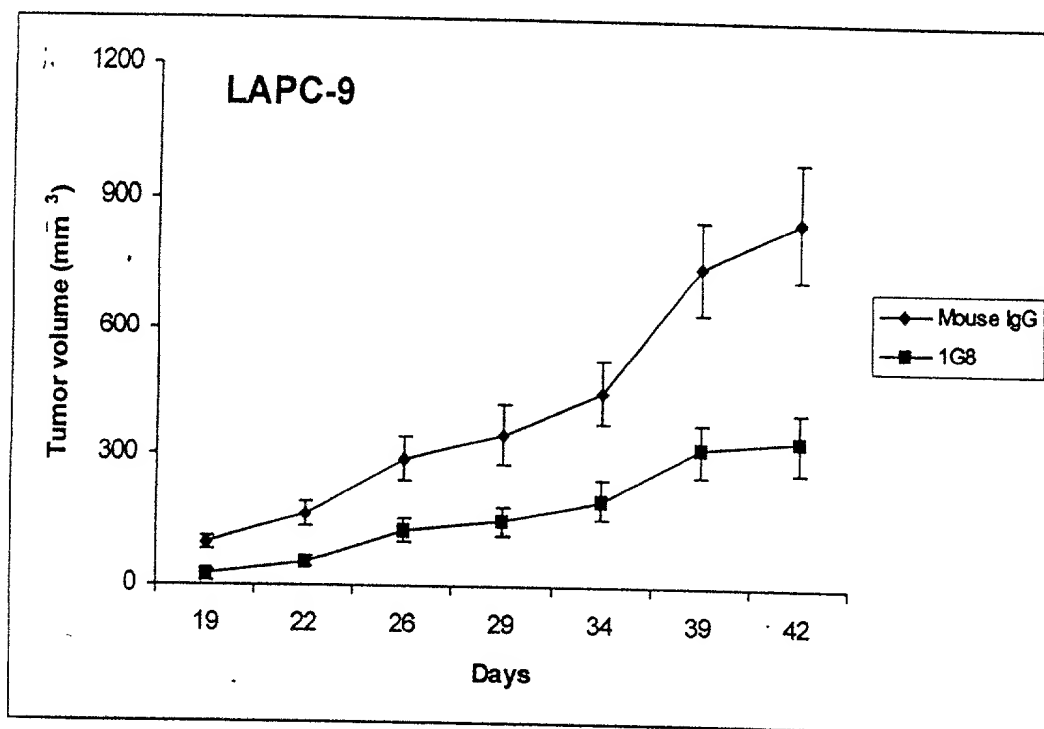
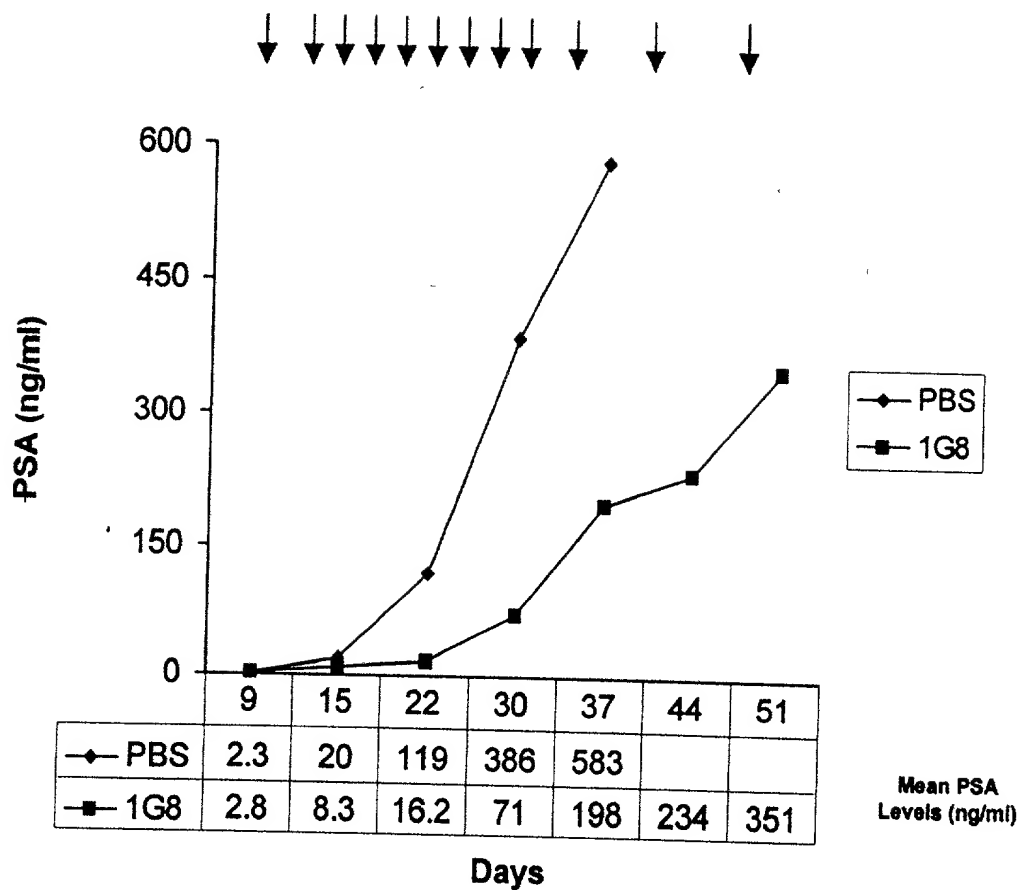


FIGURE 65

A)



B)

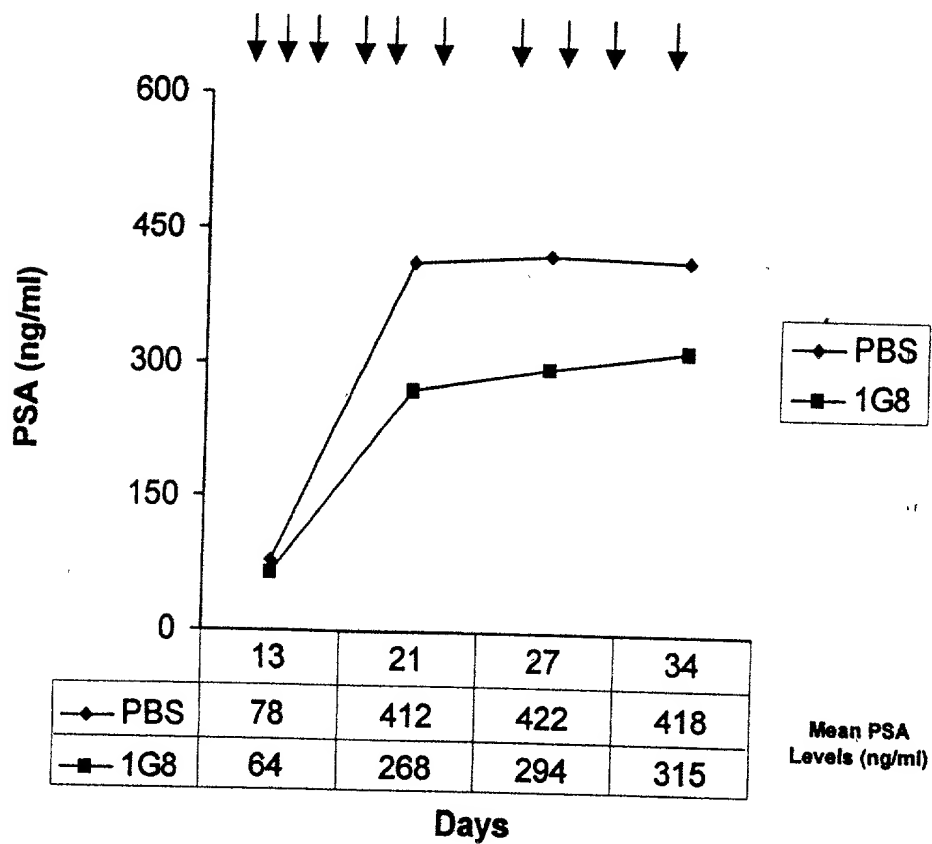
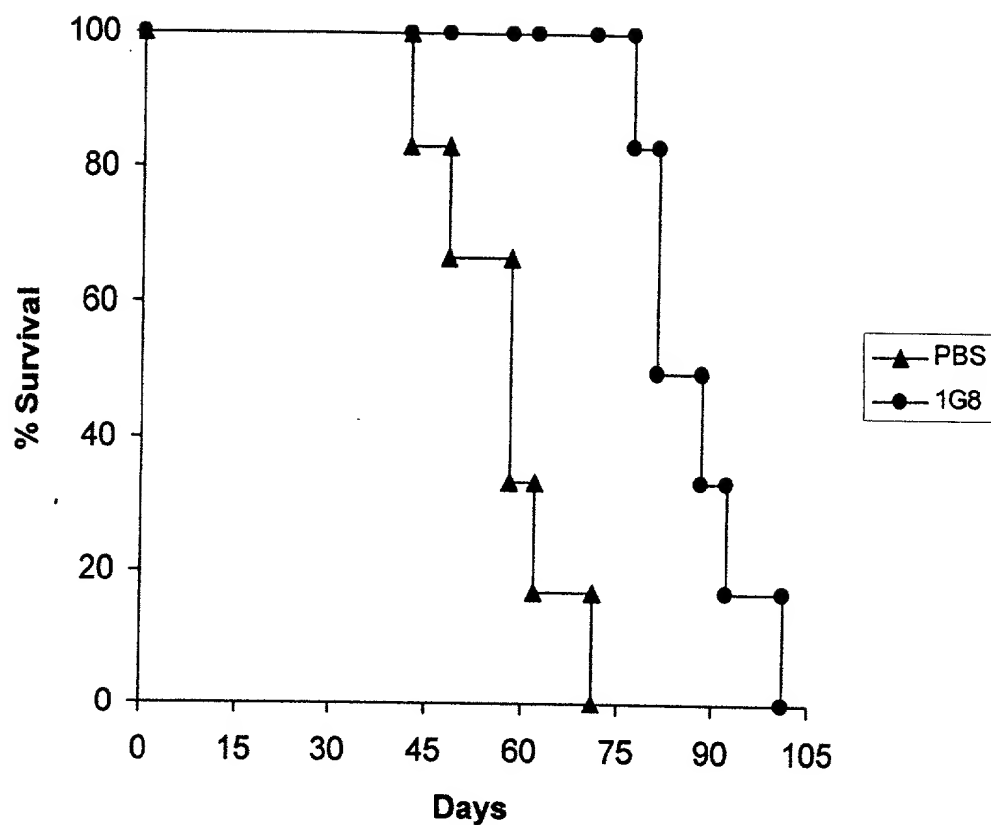


Figure 66

A)



B)

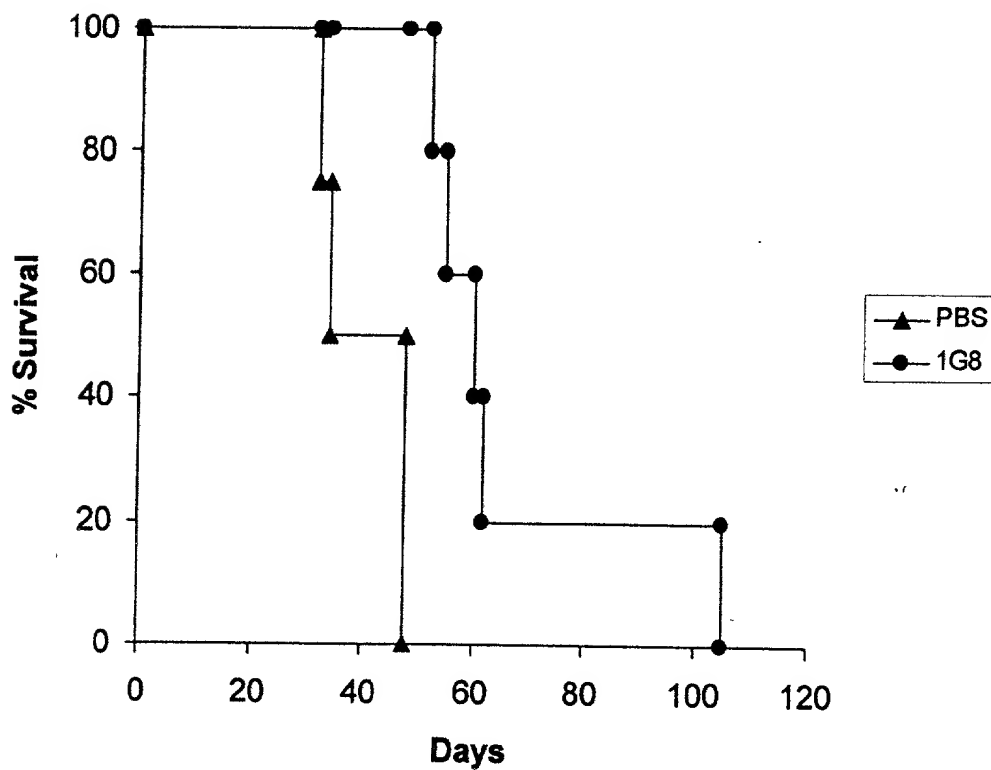
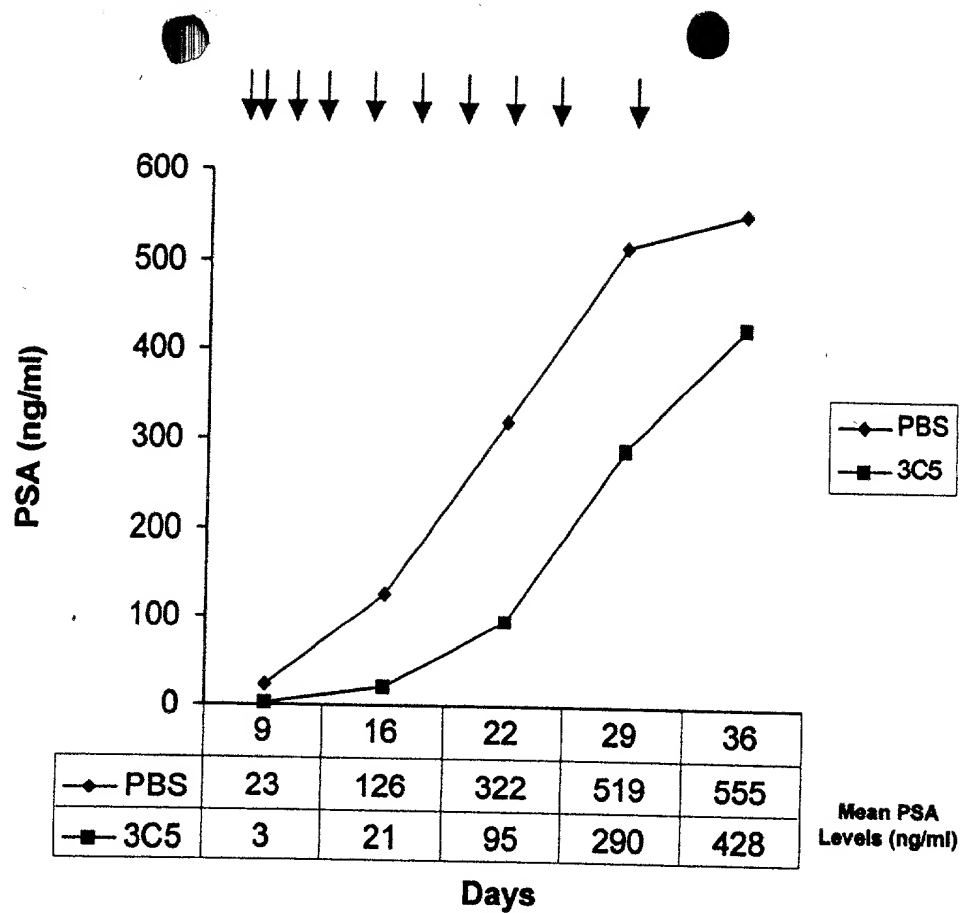


Figure 67

A)



B)

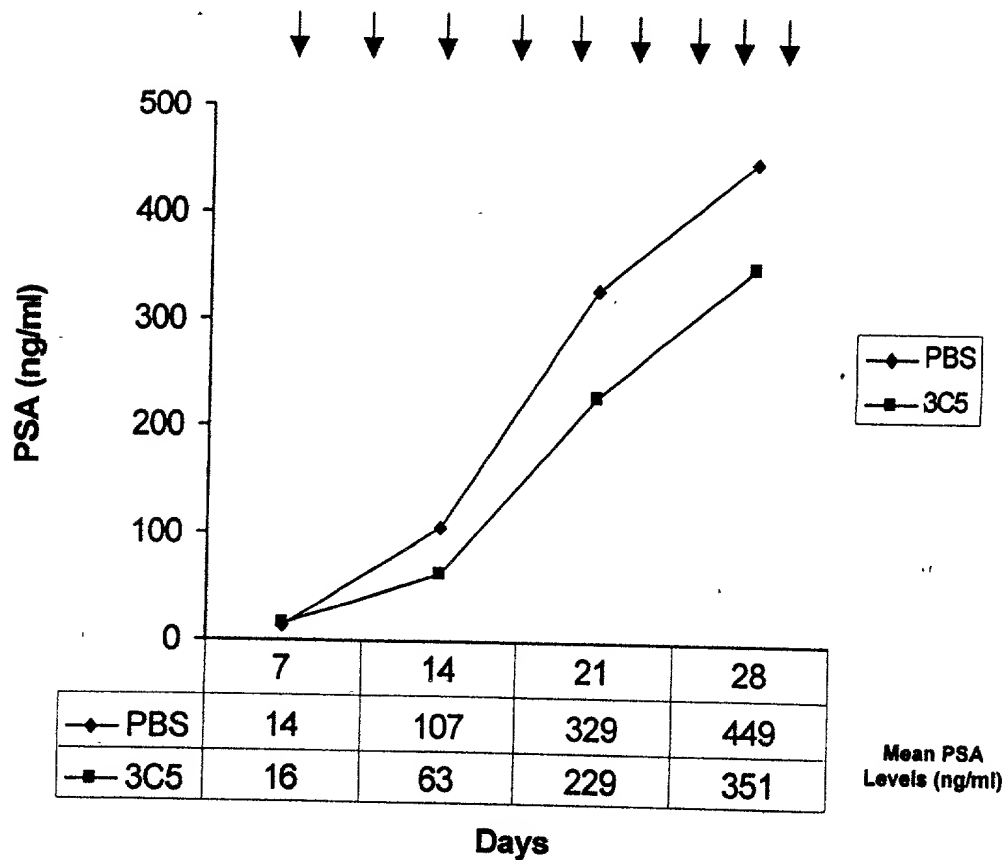
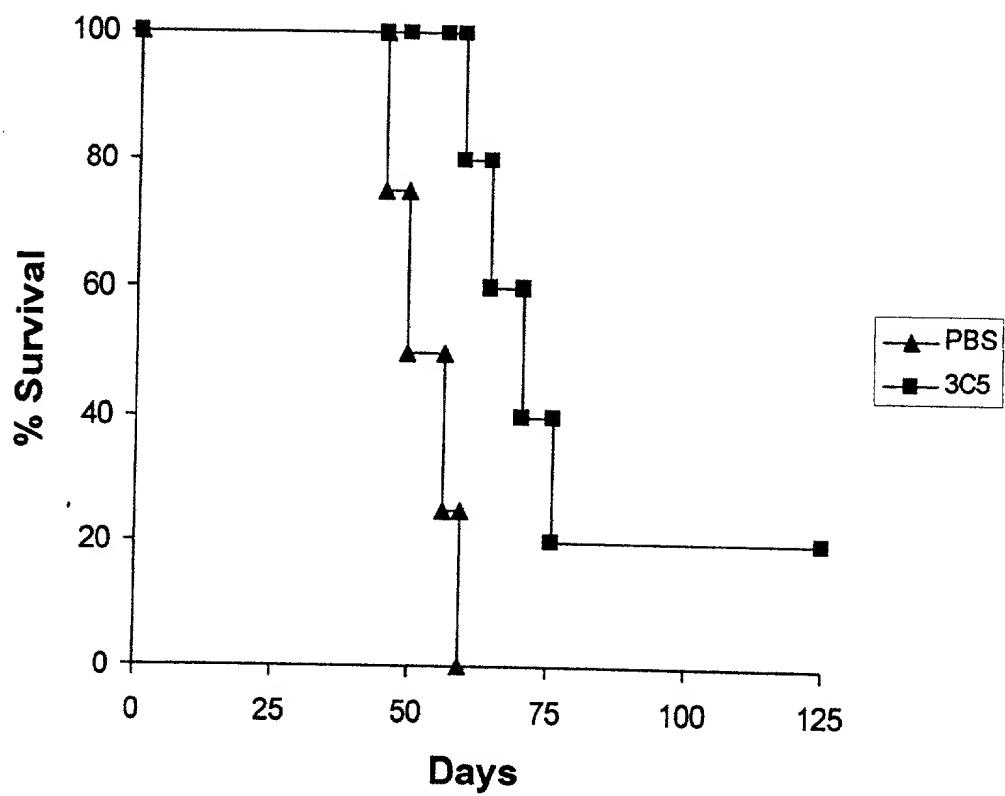


Figure 68

A)



B)

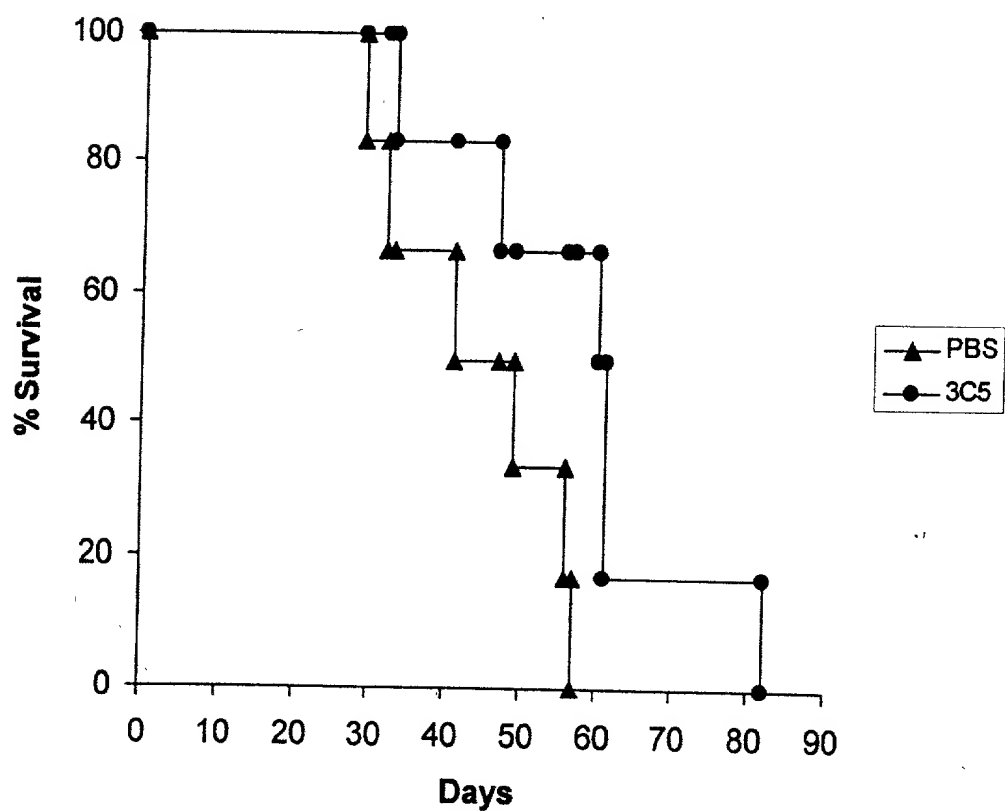


Figure 69

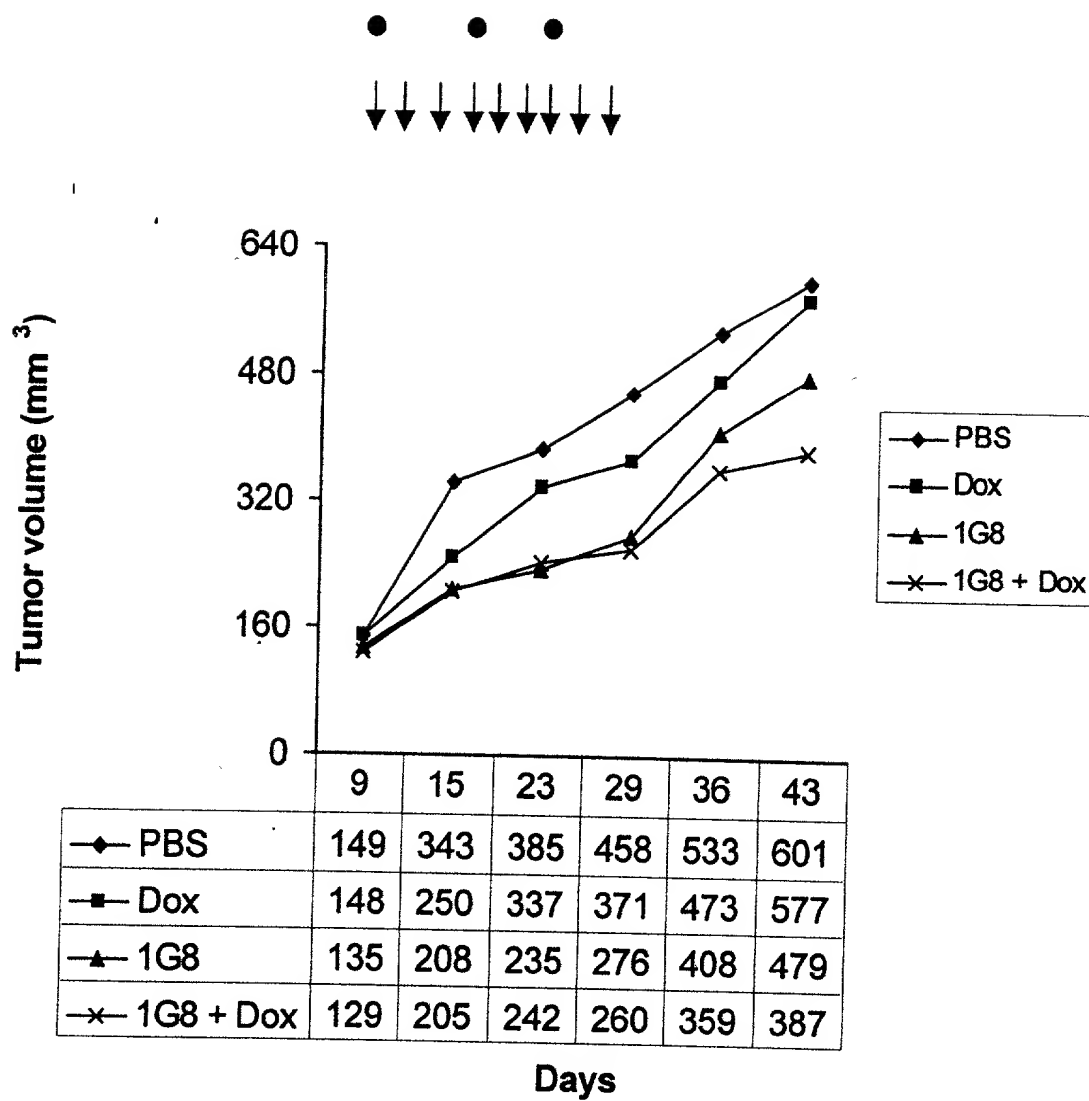
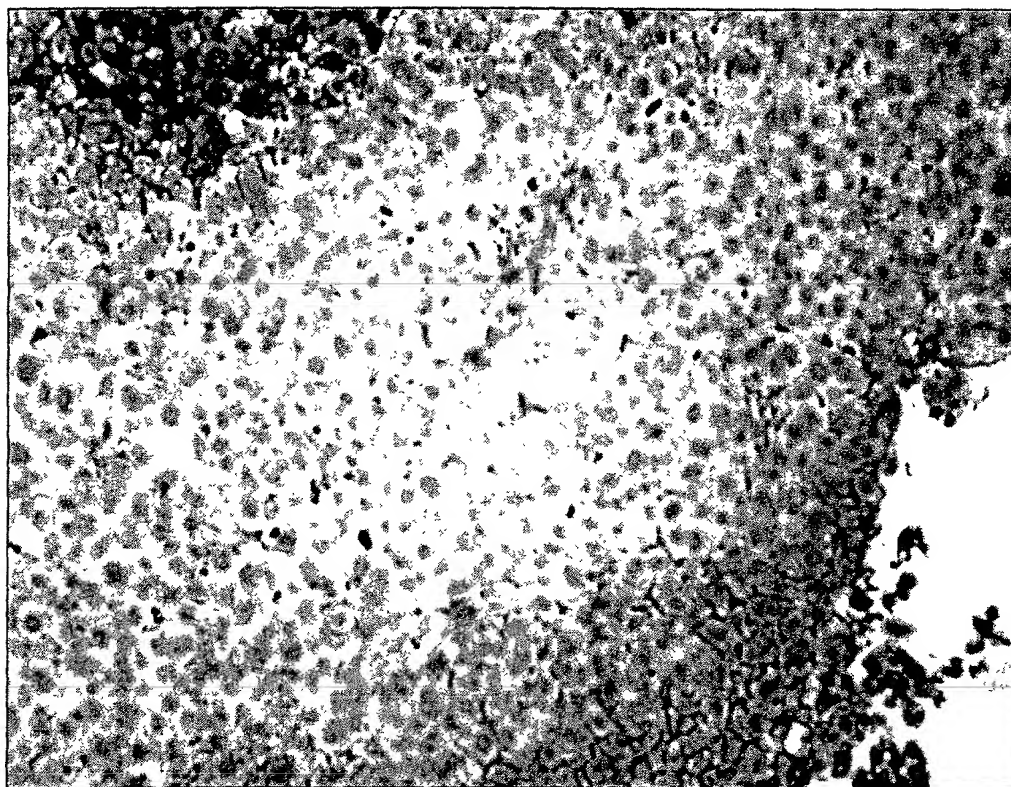


Figure 70

PSCA 3C5 MAb Localizes within LAPC9AD Xenograft Tissue

3C5 Treated



mIgG Treated

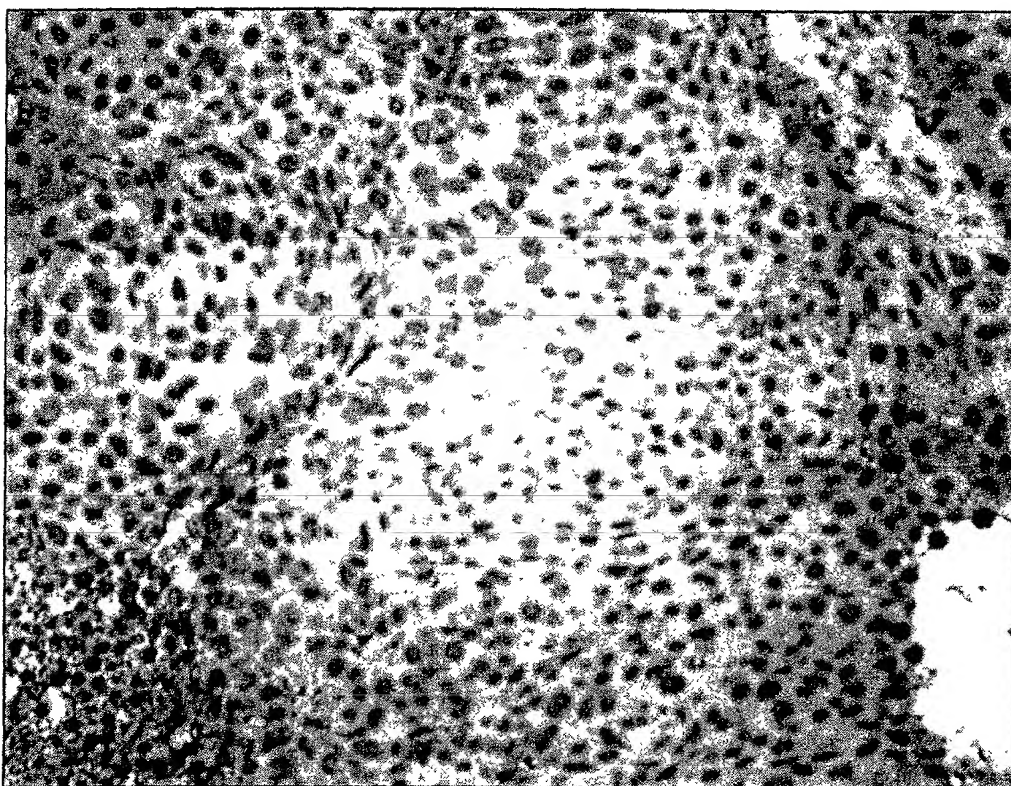


Figure 71

123456789101112131415161718192021222324252627282930313233343536373839404142434445464748495051525354555657585960616263646566676869707172737475767778798081828384858687888990919293949596979899100

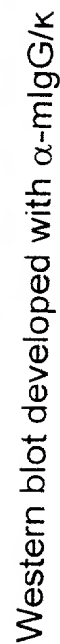
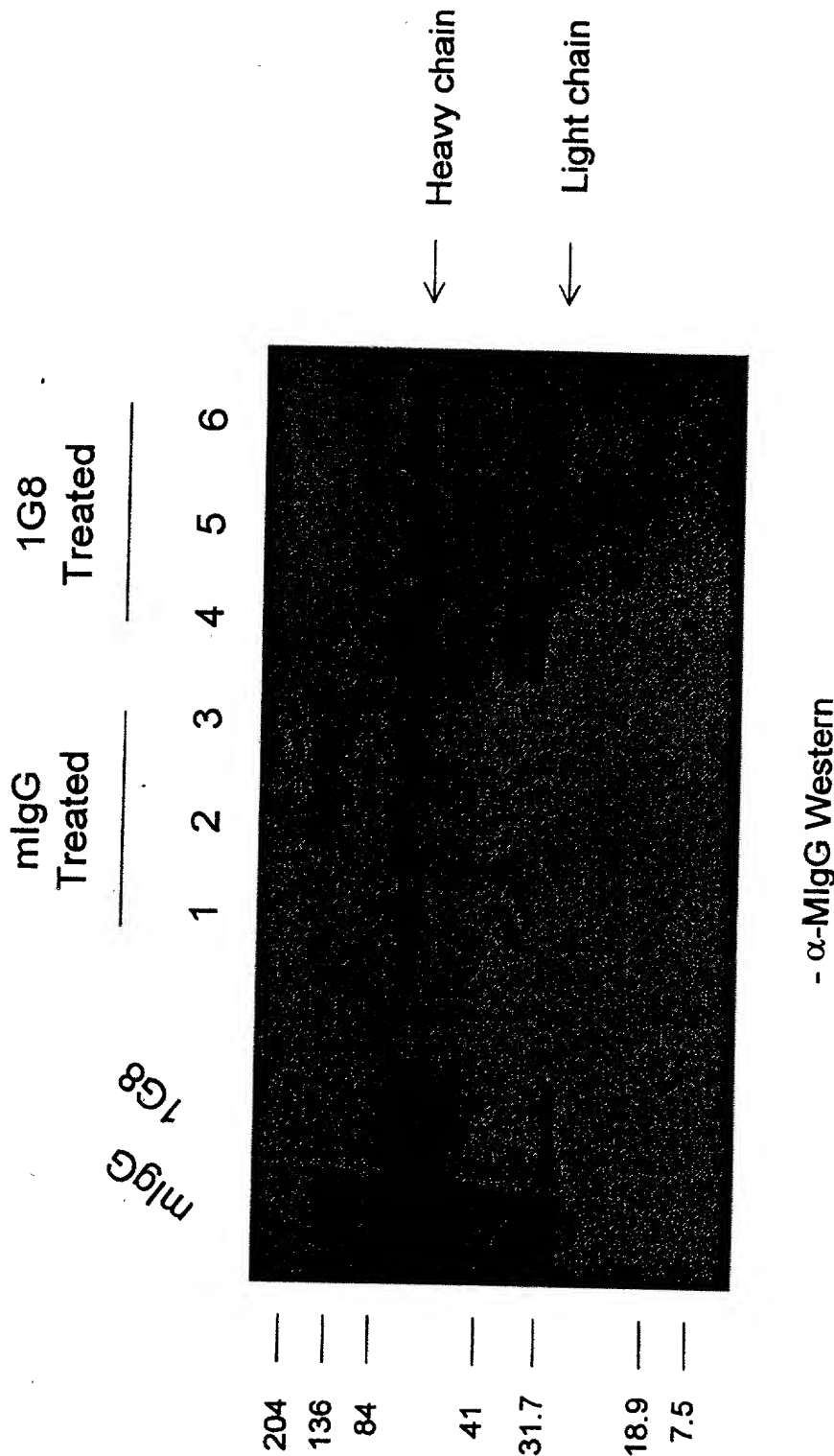


Figure 72

SPECIFIC TARGETING OF THE 1G8 ANTI-PSCA MAb TO ESTABLISHED LAPC-9 TUMORS



Method: Mice bearing established LAPC-9 tumors (>100 mm³) were injected with either mlgG or the anti-PSCA MAb 1G8. Tumors were harvested a week later and made into protein lysates for Western analysis.

Figure 73